Pollinator effectiveness and their potential for pollination of

greenhouse crops in Kenya

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"I will be a hummingbird,

doing the best I can to save the biodiversity."

Prof. Wangari Maathai (1940-2011) Environmental and political activist and Nobel Peace Laureate 2004.

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List of Abbreviations

μl	Microlitre
μm	Micrometer
€	Euros
AILVs	African Indigenous Leafy Vegetables
ANOVA	Analysis of Variance
BIOTA	Biodiversity Monitoring Transect Analysis in Africa
CBS	Central Bank of Kenya
CCD	Colony Collapse Disorder
cm	Centimeter
CVM	Contingent Valuation Method
DAP	Double Ammonium Phosphate fertilizer
ERA	Economic Agriculture Review
ESD	Equivalent Spherical Diameter
EV	Total economic value
FAO	Food and Agriculture Organization
g	Grams
GDP	Gross Domestic Product
GLM	General Linear Model
ha	Hectares
HCDA	Horticultural Development Authority
IPEV	Economic Value of pollination service
IPM	Integrated pest management
IZL	Invertebrate Zoology Laboratories
JKUAT	Jomo Kenyatta University of Agriculture and Technology
KARI	Kenya Agricultural Research Institute
Kg	Kilogram
KHDP	Kenya Horticultural Development Program
KIFCON	Kenya Indigenous Forest Conservation Program
MDG	Millennium Development Goals
ml	Milliliter
mm	Millimeter
MOA	Ministry of Agriculture
MT	Metric ton
NARL	National Agricultural Research Laboratories
NMK	National Museums of Kenya
°C	Degree Centigrade
р	Probability level of significance
PS	Pollination service
RV	Vulnerability ratio to pollinator loss
SE	Standard error of difference of means
SPSS	Statistical package developed by IBM [®] SPSS [®] Statistics (version 19)
US\$	United States Dollars
USA	United States of America
USGS	Unites States Geological Survey
WTP	Willingness to Pay

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1. SUMMARY

There is worldwide concern that insect pollinators are on the decline in both diversity and abundance, and such declines might have significant negative impacts on agricultural production and as such food and nutritional security in a number of developing countries such as Kenya. Reasons for this decline include, among others, habitat fragmentation, use of pesticide in intensive agriculture, effects of climate change, and poor public awareness on the role of pollinators in agricultural production systems. In Kenya, most crop production is small-scale based with the producers practising mixed cropping system. Despite the fact that most of these crops relying on pollinators for fruit and seed set, their pollinators remain unknown making pollinator utilization as well as conservation difficult.

The results from this study indicate that crops grown in the farmlands of Kakamega forest are pollinator limited in terms of both yield and quality. Spider plant (*Cleome gynandra* L.), and slender leaf (*Crotalaria brevidens* Benth), relies heavily on pollinators for fruit and seed set with more than 90% pollinator dependence ratio. Broad-leafed African nightshade (*Solanum scabrum*) pollinator dependence ratio is only 10%. Unrestricted visits from pollinators not only increased the seed set but also the seed quality. In *C. brevidens*, percent seed germination was low (30%) in seeds from pollinator exclusion treatments, but high (76%) in hand cross-pollination and open pollination with unrestricted pollinator visit (72%). *C. gynandra* germination was low, with hand cross-pollination at dusk having 42%. *S. scabrum* recorded high germination (80%) in all the treatments.

Pollinator effectiveness studies indicated that wild unmanaged bees were the most effective, and perhaps could be the most important in the production of most crops. For instance, spider plant was effectively pollinated by the short-tongued hawkmoths: *Hippotion eson, H. osiris, Nephele aequivalensi* and *Agrius convolvuli*. They were more frequent and had high frequencies of individuals with *C. gynandra* pollen present on their proboscises. Solitary bees, *Megachile* spp. and *Xylocopa* spp. were the most frequent floral visitors to *C. brevidens* with relative abundance of 90.9% and 8.4% respectively. *Megachile rufipes* was the most promising pollinator due to its high pollinator effectiveness (0.91). Meanwhile smaller bees belonging to the families Apidae (*Plebeina hildebrandti*) and Halictidae (*Lasioglossum* sp.) were the most important pollinators of karela

(*Momordica charantia*). Their pollinator effectiveness was 0.88 each on karela. The latter was also the most effective (3) compared to *Tetraloniella buharti* (0.9) for pollination of okra (*Abelmoschus esculentus*). Carpenter bees (*Xylocopa calens*) and honeybees (*Apis mellifera*) were important for the pollination of eggplant and broad-leafed African nightshade.

Honeybees (*Apis mellifera*) awere considerd a practical and feasible option to improve fruit set and yield of greenhouse tomatoes. Flowers visited by *A. mellifera* recorded high fruit set (80%), higher number of seeds per fruit and heavier fruits compared to no facilitated pollination. Factors that would affect their efficiency are discussed and recommendation given herein. Although, *Meliponula bocandei* and *M. feruginea* did not visit tomato flowers, their use in the pollination of other upcoming greenhouse crops such as courgettes and *Brassica* vegetable seed production should be studied. Evaluation of buzz pollinators like *Amegilla* and *Xylocopa* species for pollination of tomatoes under such enclosures will be of great interest.

The economic value of pollination service stood at US\$ 400 million while the vulnerability to pollinator loss was estimated to be 9%. Regionally, Eastern province of Kenya had the highest economic gain from pollination service (US\$ 74 million), followed by Nyanza (US\$ 61 million) and Coastal province (US\$ 48 million), but the three provinces most vulnerable to pollinator loss were Nairobi (52%), Coast (45%), and North Eastern (40%).

Generally, the results indicate that seed production of the African Indeginous leafy Vegetables (AILVs) must consider the role pollinators play in the seed set and quality. It is important therefore, that their habitats be conserved to ensure farmers benefit from their pollination provision. Other than *Apis mellifera* most of these pollinators are wild and unmanaged, and nest in soil, dry wood and stems, an ecosystem management approach is recommended for maximum pollination benefit. With the immense economic gains from pollinators, the importance of maintenance and conservation of pollinators' habitat cannot be underlooked. This study also highlighted the role of native bees in the pollination of crops and identified the most effective for future breeding, artificial nesting block development and the overall conservation of native pollinators for managed pollination.

Recommendations such as awareness campaigns for policy makers, farmers and the public, and the investments in pollination research by both private and government research institutions are made. Breeding and mass production of pollinators for managed crop pollination and improved crop productivity, livelihood improvements, and food and nutritional security should form priority future research areas.

2. GENERAL INTRODUCTION

2.1 Pollination and pollinators

Pollination is the process of pollen transfer from male flower parts (anthers) to the female flower parts (stigma) on the same flower, on another flower on the same plant or another plant of the same species at times over distances (Roubik, 1995; Lord and Russel, 2002). Fertilization of the ovules can only proceed once viable pollen has been deposited on the receptive stigma. Pollination therefore is a crucial step in the sexual reproduction of most plants. Plant species may be self-fertile or self-infertile. Self-fertile species can set fruit and seed from their own pollen (self-pollination) while self-infertile species must receive pollen from other plants of the same species (crosspollination) in order to set any fruit and seed (Free, 1993). Cereals, for example rice, barley, wheat, maize, sorghum, etc are wind pollinated. Their flower morphology is constructed such that pollen is easily transported through wind or spontaneously during flower opening onto the receptive stigma. Still, cereals such as maize have been considered to be important pollen sources for honeybees and a wide range of solitary bees (Vaissiére and Vinson, 1994). Self-fertile plant species benefit from pollen vectors' visitation and may have better fruit yield and seed quality when cross-pollinated than when self-pollinated (Free, 1993). This degree of dependence on insects for pollination depends on the structure of the flowers in relation to the pollinator and their degree of self-fertility (Free, 1993; Richards, 2001). In cross-pollination, many plants rely on animals especially insects as pollen vectors (McGregor, 1976; Free, 1993). Most animal-mediated pollination systems are considered mutual relationships where plants provide the much needed food source through their nectar, pollen, oils, and even nesting sites as rewards while the insects act as pollen vectors (Faegri and van der Pijl, 1979). During the collection of these rewards, pollen from the flower's anthers may stick to the animal body. On subsequent visits by the animal to the next flowers while searching for more rewards, pollen from its body may adhere to the stigmas of these flowers thereby effecting crosspollen movement. For many plants, insects are the main pollen vectors but several species of birds, bats and other mammals also regularly visit and pollinate flowers.

Different plants including crops tend to exhibit an array of flower morphologies, nectar, and pollen characteristics that may reflect the morphology and physiology of certain pollinator types. Based on such characteristics, Faegri and van der Pijl (1979) classified pollination syndromes of

animal pollinated flowers (blossoms) as cantharophily (beetle-pollinated), ornithophily (birdpollinated), chiropterophily (bat-pollinated), melittophily (bee-pollinated), myophily (fly-pollinated), psychophily (butterfly-pollinated) and phalaenophily (moth-pollinated among others). For example, bilaterally symmetric flowers, yellow or blue in colour with nectar guides present, relatively concentrated and moderately hidden nectar are reportedly adapted for bee pollination. On the other hand, flowers adapted to hawkmoth pollination have nocturnal anthesis, with white or faintly coloured petals, lack nectar guides but with deeply hidden nectar tubes (Faegri van der Pijl, 1979). Although, a plant may exhibit a particular pollination syndrome, a wide array of floral visitors may visit the flowers for nectar and pollen rewards (Ollerton, 1996; Waser et al., 1996). However, not all flower visitors are potential pollinators. Inouye (1980) and Roubik (1995) classified various types of visitors to plants on the basis of their behaviour in relation to pollen or nectar collection as (a) pollinators, (b) thieves referring to those visitors who obtain the reward without damaging the flower, but do not pollinate it due to a mismatch of morphologies, and (c) robbers, that is, those visitors who obtain the reward by damaging floral tissues and do not effect pollination. Bees are by far the most important pollen vectors for most plant species including a number of agricultural crops worldwide (Free, 1993).

2.2 Importance of pollinators in agriculture

Only until recently has the importance of pollinators in agriculture gained momentum (Kevan and Phillips, 2001). McGregor (1976) in review documented various crops that benefit from insect pollination. Williams (1994) assessed the pollinator needs of 264 European crops and concluded that 84% of these depended to some extend on animal pollination. In the tropics, insect pollination increases fruit and seed production in 70% of tropical crops (Roubik, 1995). A recent review on dependence of crops on pollinators worldwide showed that 87 out of the 124 leading food crops are dependent on animal pollination (Klein *et al.*, 2007). Pollinators are thus very essential for sustainable food production for the human population worldwide. In Kenya as in other developing nations, pollinators' role in crop production for most important crops is not well known (Rodger *et al.*, 2004). Available studies include, watermelon (Njoroge *et al.*, 2004), bottle gourd/dudhi (Morimoto *et al.*, 2004), tomatoes, capsicums, passion fruits, pumpkins (Kasina, 2007), eggplant (Gemill-Herren and Ochien'g, 2008), sunflower (Nderitu *et al.*, 2008), and papaya (Martins and Johnson, 2009). Elsewhere, following appreciation of the significant role pollinators play in

increasing fruit/seed set and quality, managed pollination service has become an essential factor in the production of some crops such as clover seed, tomatoes and straw berries (Delaplane and Mayer, 2000). The flagrant paucity of information on crop pollination requirement could be a limiting factor in agricultural production.

2.3 Pollinators and food security

Pollinators are important in increasing yield of horticultural crops, and pastures which are critical to the maintenance of health, nutrition, food security, and farmer's incomes. Estimations indicate that, pollinators contribute 35% of the world's crop production, and increase outputs of 87 of the leading crops worldwide (Klein et al., 2007). Eilers et al. (2011) reported that those crop plants that depend fully or partially on pollinators contain more than 90% of vitamin C, the whole quantity of Lycopene and almost full quantity of the antioxidants β -cryptoxanthin and β -tocopherol, majority of lipid, vitamin A and related carotenoids, calcium, fluoride and large portion of folic acid. Yet, there is mounting evidence that these pollinators are on the decline (Buchmann and Nabhan, 1996; Potts et al., 2010). This calls for urgent measures to address this decline if food and nutrition security are to be achieved. In the case of Kenya, intensive and diversified agricultural production is one of the strategies that would alleviate food and nutrition insecurity. It can therefore be expected that the demand for pollination service will increase as the acreage under intensive agriculture increases and new crops are introduced to meet the populations' food and nutrition needs (Aizen et al., 2008; Aizen and Harder, 2009). Pollinator decline is therefore likely to impact negatively on such efforts and lead not only to food insecurity but also to malnutrition. Deliberate efforts to conserve and manage pollinators are therefore key aspects in the achievement of increased crops yields, food security, better nutrition, and better farm incomes and should therefore be rigorously pursued.

2.4 Economic importance of pollinators in agro-ecosystems

Several assessments have been undertaken on the economic contribution of pollinators to agricultural crops. Globally, the annual contribution of pollinators to the agricultural crops has been estimated at about US\$ 200 billion (Pimentel *et al.*, 1997; Gallai *et al.*, 2009). In the USA, the economic value of honeybees as agricultural pollinator has been estimated to be between US\$ 1.6-5.7 billion per year (Southwick and Southwick, 1992), US\$ 14.6 billion in 2000 (Morse and Calderone,

2000) while in United Kingdom the value is estimated at £ 137.8 million per year on some selected crops (Carreck and Williams, 1998). Ricketts *et al.* (2004) estimated that pollination services from tropical forests contributed about 7% (US\$ 62,000) to coffee production in Costa Rica. Some of these estimates were however based largely on the value of honeybees to agro-ecosystems and do not take into consideration other pollinators such as wild bees, hawkmoths, birds, flies that actively visit and effectively pollinate some crops. Few studies exist on the economic value of pollination service for most African countries. These are for South Africa (Allsopp *et al.*, 2008) and recently for Uganda (Munyuli, 2010). In Kenya, Kasina *et al.* (2009) estimated the economic benefit from bee pollination to eight vegetable crops in Kakamega district at US\$ 3.2 million. Such monetary value estimates of pollination service, are clear facts that could help convince policy makers on the need to conserve natural habitats such as forests and form strong basis for pollination research funding considerations. Comprehensive evaluation of the economic significance of pollinators to agricultural production in Kenya is therefore worthwhile and herein recommended.

2.5 Pollinator diversity, effectiveness, efficiency and managed crop pollination

Bees, especially solitary and bumblebees, are the most important crop pollinators (McGregor, 1976; Free, 1993). This is due to their morphological and behavioural adaptations. For example, the presence of enormous body hairs, constant nectar and pollen collection for their young, enhances their ability to transfer more pollen from anthers to the stigmas of the flowers they visit (Free, 1993; Michener, 2000). Honeybees pollinate only 15% of the world's crops while bumblebees, leaf cutter bees, and other solitary bees pollinate the bulk of the remaining crops (Buchmann and Nabhan, 1996; Winfree *et al.*, 2008). Other non-bee pollinators are essential pollinators as well. In cacao (*Theobroma cacao* L.: Sterculiaceae) for example, flowers are self-infertile and are exclusively pollinated by biting midges of the genus *Forcipomyia*: Ceratopogonidae (Young, 1994). Similarly, papaya (*Carica papaya*: Caricaceae) is pollinated by the sphingid hawkmoths (*Hippotion celerio*, *Nephele comma* and *Agrius convolvuli* (Martins and Johnson, 2009). For many agricultural systems, pollinator diversity and abundance are important for improved pollination and increased fruit and seed set. Pollinator diversity may not only enhance fruit and seed set but also reduce risks of crop loss that may be associated with decline or lack of a pollinator during the crop's flowering periods (Winfree *et al.*, 2007).

A wide variety of bee species are known to be efficient and effective pollinators of many crops (Richards, 2001; Kremen et al., 2002). Apis mellifera L. is the most commonly used species in managed pollination services (McGregor.1976; Watanabe, 1994). This is because they are versatile, cheap and easy to manage and are readily brought into the field of flowering crops whenever necessary (Delaplane and Mayer, 2000). For some crops however, honeybees is not usually the most effective (Westerkamp and Gottsberger, 2000; Kremen et al., 2002). The global decline of bee pollinators especially honeybees have highlighted the risks involved on reliance on a single pollinator (Winfree, 2008) and the urgent need to evaluate other bee pollinators for managed crop pollination (Winfree et al., 2007). Farmers in Kenya seldom practise managed pollination with intention to increase crop production. For instance, in the farmlands neighbouring Kakamega Forest, farmers rely on feral bees for pollination (Kasina, 2007). In other parts of the world, however, various native bees have been evaluated for their pollination effectiveness and developed for manage crop pollination. Bumblebees e.g Bombus impatience, B. occidentalis, B. terrestris (family: Apidae) are used in greenhouse pollination of tomatoes and capsicum in the USA and Europe. Nomia, Osmia (family: Halictidae) Megachile rotundata (family: Megachilidae) are used for commercial pollination of crops like alfalfa. Other native bee species including Amegilla (Zonamegilla) holmesi Rayment (family: Apidae) are on evaluation for pollination of greenhouse tomatoes in Australia (Bell et al., 2006). Unfortunately, for most tropical crops in Africa, Kenya in particular, such data as the diversity and effectiveness of pollinators to undertake similar evalutions on crop pollination commercialisation is still lacking.

2.6 Threats to pollinator diversity and abundance

Decline of pollinators have been reported worldwide, and nearly 200 species of wild vertebrate pollinators alongside the inverterbrate pollinators may be on the verge of extinction (Buchmann and Nabhan, 1996; Kearns *et al.*, 1998; Kevan and Phillips, 2001; Potts *et al.*, 2010). Decline in pollinator diversity and abundance will negate the efforts to increase agricultural production and maintain healthy ecosystem. A number of possible factors have been suggested, including habitat fragmentation (Steffan-Dewenter and Tscharntke, 1999; Aguilar *et al.*, 2006), pesticide use (Kearns *et al.*, 1998; Kremen *et al.*, 2002) and introduction of alien plant and insect species (Memmott and Waser, 2002, Bjerknes *et al.*, 2007). Others are honeybee diseases and insects, for example, the Colony Collapse Disorder (CCD), varroa mite (*Varroa destructor*) and

Nosema ceranae fungus (Buchmann and Nabhan, 1996; Kearns *et al.*, 1998; Kevan and Phillips, 2001; Potts *et al.*, 2010). Already, varroa mite has been reported in honeybee hives in South Africa (Allsopp, 2004) and East Africa (Fazier *et al.*, 2009). Although the effect of the varroa mite on the honeybees is still unclear, it is possible that its presence could not only affect crop pollination, but would also reduce incomes generated from hive products like honey and beewax.

Natural habitats such as forests are continually under pressure from increasing demand for food production areas and urbanization by the increasing population. Clearing of forests to meet these demands, reduces pollinator nesting sites especially soil nesting bees, forage resources and instead increases the risks of extinction of both pollinators and the plants that rely on them for fruit and seed set (Rathcke and Jules, 1993; Kearns et al. 1998; Cane, 2001; Kremen et al., 2002). Plants within fragmented landscapes have been found to have reduced seed set of 50%-60% (Allen-Wardell et al., 1998). Extensive monoculture reduces nectar and pollen resources for pollinators (Kearns et al., 1998). Furthermore, inadvertent pesticides use may affect pollinator populations and behaviour. Insecticide use will kill pollinators; poison their food resources, while herbicides will wipe off plants that act as nectar and pollen sources (Johansen and Mayer, 1990; Buchmann and Nabhan, 1996; Kearns et al. 1998; Richards, 2001). Fenitrothion insecticide use was directly linked to reduced pollinator diversity and abundance and the corresponding reductions in fruit and seed set in blueberry in New Brunswick, Canada (Kevan and Plowright, 1995). Invasive plants and insects may displace native nectar and pollen resources and result into competition for nectar and pollen resource with the native species while the latter may cause proliferation of weeds (Stout et al., 2002). Introductions of Bombus terrestris into Israel for example, negatively affected the populations of native bee species especially Apis mellifera because of competition for food resources (Dafni, 1998). Although studies on the effct of climate change on pollinators are still scarce, a recent review by Kjøhl et al. (2011) indicated that, climate change might severely affect pollinators and result into poor crop yield. Increases in temperature and reduced rainfall may cause both temporal and spatial mismatches between pollinators and crops; plants may experience reduced insect visitations while pollinators may lack the nectar and pollen resources (Memmott et al., 2007; Hegland et al., 2009).

Due to the significant role played by pollinators in agro ecosystems, such reductions in the diversity and abundance of bee species leads to reduced crop yields and quality (Potts *et al.*, 2010) e.g passion fruits in Central America (Roubik, 1995) and watermelon in the USA (Kremen *et al.*, 2002). With the increasing dependence of crops on pollinators parallel to the global trend of decline

of pollinators (Aizen *et al.*, 2008), crop productivity and food security may be threatened (Allen-Wardell *et al.*, 1998). It is therefore important to identify these crops' pollinators, their food resource and nesting requirements to enable better management and policy formulations that would sustainably maintain their populations, increase crop production, and maintain healthy ecosystems.

2.7 Problem statement

Very little information exists on the pollination of many horticultural crops in Africa (Rodger *et al.*, 2004). In Kenya, for example, only a few studies are available to demonstrate that pollinators are essential for fruit and seed set, consequently yield of many crops. But, for some crops, even in the presence of high fertilizer use little or no yield will be realised without pollinators. The study by Njoroge *et al.* (2004), revealed that watermelon (*Citrullus lanatus* Thunb. Cucurbitaceae) relies heavily on pollinators for fruit set and quality. Nderitu *et al.* (2008) indicated that pollinators not only influenced the seed set but also the oil quantity in sunflower (*Helianthus annuus* L.: Asteraceae). Kasina (2007) found out that crops grown in the farmlands of Kakamega Forest e.g passion fruits and squash depended heavily on pollinators for fruit set. Whenever pollination studies exist, comparison on pollination effectiveness among the floral visitors is often lacking. For African Indigenous Leafy Vegetables (AILVs), no information on pollination or pollinators exists. More information on the role of pollinators in crop production is necessary for better policy formulations and conservation strategies.

In addition, the increasing shift from open field fruit and vegetable production to greenhouses, present an increased need for managed crop pollination. These conditions exclude insects such as bees, and fruit set and quality is low probably due to insufficient pollination. There are no reports of managed pollination under such conditions in Kenya. In fact, farmers when considering crops to grow under greenhouse conditions rarely consider the pollination needs of the crops. For example, high pollinator dependent crops like watermelon or courgette are grown under these enclosures without any pollinator. Consequently, even after such a high investment endeavour, farmers still record losses due to flowers not setting fruits. Reasons for this include lack of public awareness on pollination issues, lack of information on the effective pollinators and their nesting habits and their possible management for enhanced crop pollination. Introduction of new crops like vanilla requires the government's investment in screening of the most efficient pollinator amongst

the array of native bee pollinators we have. Its pollinator, the stingless bee *Melipona* is absent in Kenya and its fruit production exclusively relies on hand pollination which forms the bulk of its production costs. Furthermore, with the increasing acreages under agricultural production coupled with the global decline of pollinators, the demand in pollination and pollinators will soon be overwhelming. It is therefore necessary to document these pollinators and evaluate their effectiveness for future breeding and utilization in managed pollination. Finally, estimations for the economic value of pollinators and invoke positive reactions in terms of willingness of the government to fund pollination research studies and to formulate supportive pollinator conservation policies for sustainable agricultural production and ecosystem maintainence.

2.8 Study Area: Kakamega forest farmlands

The study was conducted within the farmlands around Kakamega forest located between latitudes 00° 08' 30.5"N (41 236 in UTM 36 N) and 00° 22'12.5"N (15 984) and longitudes 34° 46' 08.0" (696 777) and 34° 57' 26.5" E (717 761) and altitude of about 1,500 to 1,700 m above sea level (KIFCON, 1994) (see Fig.2.1). The farmlands of Kakamega forest consist of rich agricultural soils, and the high rainfall of about 2,000 mm is well distributed through the year. Peak rainfall occurs in April-May (long rains) and October-November (short rains) with mean monthly temperatures range from 11°C to 29°C with an average daily temperature of 22°C (Jaetzold and Schmidt, 1982). The forest is an important habitat for a large number of rare animal and plant species, some of which are endemic (KIFCON, 1994). It also offers a range of ecosystem services that are rendered to farmlands by wild flora and fauna, including pollination, seed dispersal, nutrient cycling, and soil conservation (BIOTA, 2004). This rich biodiversity is at jeopardy due to habitat fragmentation, mainly due to human encroachment for agriculture, leading to degradation of natural habitat and its biodiversity. Agricultural production in the farmland area is characterised mainly by small scale farming with small land units of 0.2 ha to 0.7 ha per household (MOA, 2010). Sugarcane is the most dominant cash crop while other crops include maize, beans, pumpkins, vegetables such as *Brassica* spp. and African Indigenous Vegetables (AILVs) for example, Cleome gynandra, Crotalaria brevidens and Solanum scabrum and fruits that are mainly for the farmers' household food requirements. Beekeeping in Kakamega District is conducted on a small-scale basis only, without high commercial intention. Farmers construct hives, but do not rear queens or colonies, thus rely on feral honeybee

colonies to enter the hives (Hagen and Kraemer, 2010). Most of the crops grown rely either on these feral bees or the many solitary bees for pollination and would be strongly affected by any decline in the pollination service (Kasina, 2007).

Gikungu (2006) recorded more than 234 bee species visiting flowers in the forest and the adjacent farmlands. Some of these bee species, for example *Xylocopa calens*, depended on the forest for their nesting site and pollinated important crops such as passion fruits in the farmland region (Kasina, 2007). The farmland landscapes consist of small land units with diverse hedgerows, field margins with diverse flowering plants that would play an important role in providing food resources (nectar and pollen) for various species of bee pollinators (Mwangi, 2009). For sufficient bee pollination, management of these landscapes is important (Kasina, 2007).



Figure 2.1: Map of Kenya showing location of Kakamega forest. Landsat ETM+ (7) satellite image (5th February 2001, spectral bands 5/4/3, contrast enhanced) of Kakamega forest and its peripheral fragments (Source: USGS; access and preparation: BIOTA-E02, G. Schaab, Karlsruhe, Germany).

2.9 Justification of the study

Poverty and hunger (food and nutritional insecurity) alleviation are some of the development goals set for developing nations by the United Nations Development Programs inorder to achieve the Millenium Development Goals (MDGs) by the year 2015 (UN Millenium Project, 2005). Intensive agricultural production is one of the ways of ensuring food security and nutrition. However, this may only be achieved if pollinators are available both in abundance and diversity. This is true for crops that rely heavily on pollinators for fruit set for example, fruits and vegetables. Research scientist, breeders, and policy makers alike have ignored pollination needs of most of these crops. For instance, pollination needs of some important AILVs such as spider plant (Cleome gynandra L.: Cleomaceae), slender leaf (Crotalaria brevidens Benth: Fabaceae) and broad-leafed African nightshade (Solanum scabrum Mill: Solanaceae) are still unknown. This is unlike most vegetables whose pollination needs are contained in the review by McGregor (1976). Contrasting to reports that crop yield may be compromised without pollinators, efforts aimed at increasing food production have focused more on other inputs of production such as water, fertilizers, and crop protection against insect pests and diseases. To improve production of these crops, it is important that their pollinators be identified and conservation strategies formulated for the sustainable management of the pollinators. Already, there are global concerns that pollinator are on the decline. This study therefore aimed to provide more information on the pollination and pollinators of various vegetable crops including the AILVs and document their effectiveness on the fruit and seed set of the vegetable crops. The results will be of importance and contribute to the knowledge on pollination and pollinators of crops in Kenya.

2.10 Study objectives

The overall objective was to document the pollinator diversity range for the study crops, their effectiveness, and potential in the pollination of greenhouse crops in Kenya. To achieve this, several specific objectives were formulated.

- Describe the floral morphology and pollination needs of the African Indigenious Leafy Vegetables (AILVs) and evaluate the role of pollination on the quality of seed for the AILVs.
- 2. To assess the diversity and effectiveness of pollinators of selected vegetable crops grown in the farmland neighbouring Kakamega Forest, Western Kenya.

- 3. To evaluate the potential of different bee species for pollination of greenhouse tomatoes in Kenya.
- 4. Undertake a national economic analysis of the contribution of pollinators to agricultural production.

2.11 Research questions

- 1. Do the AILVs flower morphologies hint at any specialised pollination?
- 2. Do fruit set, seed set and quality of the AILVs depend on insect pollination?
- 3. How does the distance from the forest edge influence fruit and seed set for spider plant (*Cleome gynandra*)?
- 4. Are vegetable crops in the farmlands neighbouring Kakamega forest pollen limited?
- 5. Who are the pollinators of these crops?
- 6. Which of the pollinators is most effective and efficient in the pollination of these crops?
- 7. How do stingless bees species; *Meliponula bocandei*, *M. ferruginea* and honeybees (*Apis mellifera* L.) compare in foraging behaviour on tomatoes flowers under greenhouse conditions?
- 8. Does temperature and humidity affect bee behaviour and foraging under greenhouse conditions and if so, how?
- 9. Which of the bee species has potential as alternative to the current manual vibration of tomatoes for better fruit set?
- 10. What is the effect of supplementary pollination using different bee species on fruit set and quality?
- 11. What is the value of pollinators to agricultural production in Kenya and what are the future research and management implications?

2.12 Thesis structure

The first chapter of this thesis gives an overall summary of the study, highlighting the key findings. This is followed by a review of literature on the area of the study given in chapter 2. In the third chapter, I focused on the pollination needs of selected African Indigenous Leafy Vegetables

(AILVs). Here, AILVs refer to, spider plant (*Cleome gynandra* L.: Cleomaceae), slender leaf (*Crotalaria brevidens* Benth: Fabaceae), broad-leafed African nightshade (*Solanum scabrum* Mill: Solanaceae). Their different pollination syndromes as depicted from the floral morphology as well as the influence of pollination on quality of the seeds are elucidated. The influence of the location of seed production sites in relation to distance from natural habitats such as forests was also investigated.

The fourth chapter, focused on the diversity of floral visitors of horticultural crops including the AILVs grown around Kakamega forest. Experiments were further performed to compare the effectiveness of these floral visitors on the seed set of the test crops. This would be necessary for future study of nesting biologies, mass culture, and management of the effective pollinators for enhanced crop production.

The fifth chapter therefore, focused on the evaluation of the potential of three native bee species: (1) honeybee (*Apis mellifera*) Hymenoptera: Apidae), two stingless bees (2) *Meliponula bocandei* (Hymenoptera: Apidae) and (3) *Meliponula ferruginea* (Hymenoptera: Apidae) for the pollination of greenhouse tomatoes.

Despite evidence that for most crops yield increases in the presence of pollinators, crop pollination research and pollinator conservation are hardly considered. In order to be able to convince farmers, the public and policy makers on the importance of conservation of pollinators as well the need of investment in research and development in pollination studies, a national economic evaluation for the contribution of pollination service to Kenya's Agriculture was performed in chapter six of this study.

3. POLLINATION NEEDS AND SEED QUALITY OF SELECTED AFRICAN INDIGENOUS LEAFY VEGETABLES IN KAKAMEGA FOREST FARMLANDS

3.1 Introduction

African Indigenous Leafy Vegetables (AILVs) are important source of nutritious foods and form one of the main components of the household daily diets both in the rural and urban areas of Kenya. The leaves and tender shoots are consumed cooked in meals. They contain high levels of beta-carotene, vitamin C and moderate levels of calcium, magnesium, and iron micronutrients (Mnzava, 1986; 1990; Maundu *et al.*, 1999; Abukutsa-Onyango, 2003; Lyimo *et al.*, 2003) (Table 3.1). They play a significant role in ensuring food and nutrition security for both rural and urban populations. They are also alternative sources of income generation for the subsistence farmers and contribute significantly to the horticultural sector. A total of 158,000 metric tons worth about US\$40 million of AILVs fresh leaves were produced in 2009 (MOA, 2010). Spider plant (*Cleome gynandra* L.: Cleomaceae), slender leaf (*Crotalaria brevidens* Benth: Fabaceae), and broad-leafed African nightshade (*Solanum scabrum* Mill: Solanaceae) are some of the most important of AILVs species in Kenya (Chweya and Mnzava, 1997; Schippers, 2000).

Vegetable name	Vitamin C	Protein	Crude fibre	Fat	Calcium	Iron
	(mg/100g)	(%)	(%)	(%)	(mg/100g)	(mg/100g)
Vine spinach (Bacella alba)	98.7	5.0	1.5	0.7	250.0	4.0
Nightshade (Solanum nigrum)	234.5	1.0	0.8	0.2	66.8	2.5
Spider plant (Cleome gynandra)	89.6	1.5	0.8	0.3	40.5	0.8
Amaranth (Amaranthus spinosus)	249.0	4.6	1.6	0.6	43.2	3.8
Amaranth (Amaranthus hybridus)	58.1	4.8	1.5	0.6	246.8	2.9
Jute mallow (Corchorus sp.)	143.9	4.2	1.9	0.7	112.1	4.0

Table 3.1: Nutrient composition of dried leaves of selected African Indigenous Leafy Vegetables

Slender leaf (Crotalaria brevidens) and cowpea leaves (Vigna unguiculata) not included. Adapted from Lyimo et al. (2003).

3.1.1 Spider plant (Cleome gynandra L.: Cleomaceae)

Spider plant (*Cleome gynandra* L. syn. *Gynadropsis gynandra* L. Briq.), also known as 'cats whiskers' was until recently placed in the family Capparaceae but was later assigned to its own family, the Cleomaceae which consists of about 180-200 species most of them occuring in the warmer regions of the world (Hall *et al.*, 2002; Sanchez-Acebo, 2005). *Cleome* is native to the tropical Africa and Central America (Chweya and Mnzava, 1997; Schippers, 2000). In Kenya, *C. gynandra* is utilized as a leafy vegetable. Normally, young shoots and leaves are sold in bundles in the grocery market and supermarkets. More recently, *C. gynandra* has been reported to have insecticidal, anti-feedant and repellent characteristics for the control of aphids, thrips, diamondback moth and red spider mite populations in greenhouse production systems (Nyalala and Grout, 2007).

Many *Cleome* species are protandrous, requiring cross-pollination (Chweya and Mnzava, 1997). Self compatibility has been shown in *Cleome afrospina* (Iltis, 1967). Both self and cross-pollination have been shown to occur in *C. gynandra* (Omondi, 1990). In the same study, he observed that the high rate of out-crossing could have been a result of high phenotypic variability and the fact that anthers dehisce when flowers have been opened for a long time and their stigmas exposed and suggested that honeybees, spiders and perhaps even wind could be potential pollinators. The time of anther dehiscence, stigma receptivity and the flower morphology was not mentioned.

Flower morphology of *C. gynandra* reveals a "phalaenophily" pollination syndrome as described in Faegri and van der Pijl (1979). The flowers are faintly coloured or white with nocturnal anthesis and produce conspicuous amounts of dilute nectar. In South Africa *C. gynandra* is reportedly pollinated by unidentified ants (Makgakga, 2004). It is likely that the ants collected nectar only and were not responsible for any pollination. A study on *Cleome spinosa* in Brazil by Machado *et al.* (2006), recorded glossophagine (*Glossophaga soricina*) and phyllostomine (*Phyllostomus discolour*) bats as the major pollinators while sphingid moths *Agrius cingulata* and *Erinnyis ello* were considered as nectar robbers. Nectar collecting bats are absent in Kakamega and sphingid moths could be the most likely pollinator. In another study on *Cleome lutea* and *C. serrulata*, Cane (2008) indicated the flowers to be receptive nocturnally, but diurnal floral visitors *Apis mellifera, Megachile rotundata*, wasps, butterflies were credited as pollinators but ironically, no nocturnal visitors were observed. Mnzava (1986) related the low seed yield in Zambia with intermittent sterility, indeterminate flowering and consequent seed shattering and a pest complex that caused poor silique

set and seed set. Physiological maturity, temperature and light have also been reported to have influence on germination of *C. gynandra* (Ochuodho and Modi, 2005, Ekpong, 2009). This study therefore describes the pollination needs of *C. gynandra* and evaluates the effect of pollination on the seed set and quality. In addition, other factors such as increased distance from natural habitats is known to reduce fruit set and quality of hawkmoth pollinated crops e.g papaya (Martins and Johnson, 2009). It could be possible that the location of the seed production site from a natural habitat such a forest would greatly influence seed set of other hawkmoth pollinated crops. To verify this, a transect study was undertaken to find out if the distance from the natural habitat such as forest would influence the resultant seed set of *C. gynandra*. This is essential as habitat management is a practical means to encourage the abundance of such wild pollinators in the farmlands.

3.1.2 Slender leaf (*Crotalaria brevidens* **Benth: Fabaceae**)

Slender leaf or Ethiopian rattlebox (*Crotalaria brevidens* Benth syn. *Crotalaria intermedia* Kotschy) is one of the most important African Indigenous Leafy Vegetables (AILVs) (Chweya and Mnzava, 1997; Schippers, 2000; Abukutsa-Onyango, 2003). *Crotalaria* is native to Africa with over 400 species found in Eastern and Southern Africa (Polhill, 1982). Two of these species are commonly referred to as slender leaf: *C. brevidens* and *C. ochroleuca* are utilised as vegetables (Schippers, 2000; Abukutsa-Onyango, 2003). Other than its high nutritive value, other uses include nitrogen fixing in intercrop, as companion crop for suppressing root-knot nematodes and the suicidal germination of *Striga*, a major weed in maize and millet production systems (Schippers, 2000; 2002; Abukutsa-Onyango, 2004).

Breeding experiments have demonstrated that plants in the genus *Crotalaria* are self compatible (Etcheverry, *et al.*, 2003; Jacobi *et al.*, 2005) and capable of spontaneous self-pollination (Endress, 1996). However, spontaneous self-pollination does not occur unless the stigmatic surface has been stimulated by specific pollinators that combine strength and behaviour to expose the reproductive structures (Free, 1993). In a review of crop pollination and pollinators, McGregor (1976) noted that lack of pollinators probably helped to explain why in some parts of India few flowers of *Crotalaria juncea* set seed. Also, Nogueira-Couto *et al.* (1992) in Orwa *et al.* (2009) reported that in Brazil when pollinators were absent, *C. juncea* did not produce any pods. Usually

pollen is concealed and presented secondarily when legitimate pollinators, typically large bees strong enough to depress the keel, expose the stigma and push out a mass of pollen grains (Endress, 1996; Westerkamp, 1997). Several species both from medium to large bee species have been recorded visiting other *Crotalaria* sp. These include; *Xylocopa frontalis* and *X. grisescens* on *Crotalaria juncea* (Nogueira-Couto *et al.*, 1992 in Orwa *et al.*, 2009) and *C. retusa*, *C. pallida* and *C. lanceolata* (Jacobi *et al.*, 2005), *Epanthidium erythrocephalum* and *Pseudocentron* sp. (Megachilidae) on *C. stipularia* (Etcheverry, 2001). McGregor (1976) pointed out that many authors found *Apis mellifera* to be good pollinators of *Crotalaria* spp. Nogueira-Couto *et al.* (1992) in Orwa *et al.* (2009) indicated the need for pollinators in *Crotalaria*; when pollinators were absent the plant did not produce pods. While a lot of work has been done to demonstrate the importance of pollinators in other *Crotalaria* species, little research has been done on the pollination biology and ecology of *C. brevidens.* This is despite the fact that such information is essential if effective breeding and increased crop production is to be realized in the species. This study aims at providing information which could contribute to the better understanding of the pollination biology and ecology of C. brevidens.

3.1.3 Broad-leafed African nightshade (Solanum scabrum Mill: Solanaceae)

Broad-leafed African nightshade (*Solanum scabrum* Mill) belongs to the Solanaceae family. It is a common vegetable in the lowlands and highlands of West and East Africa but with a wide range diversity occurring in the warm humid belt of West and Central Africa (Schippers, 2000). Other uses include herbal medicine using leaf extracts as treatment for various stomach infections including stomach ulcers and stomach-ache and fodder for cattle and goats (Schippers, 2000). Flowers of *S. scabrum* are mainly self-pollinating but with low level of out-crossing (Fontem and Schippers, 2004).

Solanaceous flowers only offer pollen and not nectar to floral visitors (Mc Gregor, 1976; Free, 1993). Usually, pollen is contained in sac-like poricidal anthers characteristic of buzz-pollination and pollen can only be removed from the anthers by thoracic muscle vibration of bees (Buchmann, 1983). Depending on the flower morphology, spontaneous self-pollination may be favoured in short-styled flowers (Free, 1993). On the other hand, in the case of long-styled flowers,

more cross-pollen is likely to be deposited on the stigma due to contact with the floral visitor's body. Fontem and Schippers (2004) reported the flowers of *S. scabrum* to have long styles (3-4.5 mm) with relatively short anthers (2-3 mm). It is speculated that floral visitors may improve fruit and seed set. Floral visitors so far recorded include honeybees, and bumblebees and black syrphid flies. However, to date, no studies have investigated the pollination needs of this vegetable. Low seed germination has been attributed to low vigor due to improper seed extraction (Schippers, 2000). This study will investigate the role of pollinators on the seed set and quality of this vegetable.

Despite the fact that AILVs are integral part of agricultural systems and food diets, most African countries have not given them priority in crop development. Lack of seeds, poor and delayed seed germination are some of the production bottlenecks (Chweya and Mnzava, 1997, Abukutsa-Onyango, 2007). In the recent past, consumers have become increasingly aware of the nutritional and medicinal value of AILVs. This has led to a rise in demand especially in major urban centres, however, the supply of these vegetables is far too low to meet this growing demand. Since commercial seed production systems is undeveloped for these crops in Kenya, growers rely on seed collection from fruiting plants. In addition, there has been no research to understand reproductive needs for these AILVs. This study was therefore carried out to determine the pollination needs of these AILVs and measured their degree of dependency on pollen vectors for fruit set, seed quality and reproductive success.

3.2 Materials and Methods

3.2.1. Study site and test crop planting

The research study was carried out within the farmlands surrounding Kakamega forest in Kenya. The experiment was conducted during the long rains season (April-May) in 2009 and 2010. Seeds of three African Indigenous Leafy Vegetables, (1) spider plant (*Cleome gynandra* L.: Cleomaceae), (2) slender leaf (*Crotalaria brevidens* Benth: Fabaceae), and (3) broad-leafed African nightshade (*Solanum scabrum* Mill: Solanaceae) were collected from Jomo Kenyatta University of Agriculture and Technology (JKUAT). The choice of this source of seed was influenced by the fact

that the Horticulture Department of JKUAT is responsible for the production of quality seeds for the conservation of AILVs as one of its focus areas. A spacing of 60 cm by 120 cm in a 10 m by 10 m field was used for all the AILVs. At planting, DAP fertilizer was applied at a rate of 100 kg ha⁻¹, each plot received 1 kg of DAP. Seeds of *C. brevidens* were sown on the 4th April 2009 while *S. scabrum* on the 15th April 2009 and *C. gynandra* on the 31st March 2010. Two to three seeds were planted per hole but later thinned to one plant per hole after seedling emergence. Weeding was done every three weeks using hand held hoes. Insect pests and diseases were controlled using appropriate insecticides and fungicides whenever necessary. These sprayings were done late in the evening at the end of the week outside data collection days. Flowering first occurred in *C. brevidens* on 10th June 2009, in *S. scabrum* on 17th June 2009 and in *C. gynandra* 11th May 2010.

3.2.2 Flower morphology, nectar standing crop, concentration and pollination needs of African Indigenous Leafy Vegetables

Flower morphology, nectar standing crop and concentration

In each case, days to flowering and the duration during which the flowers remain open were recorded. Flower morphology in relation to pollination was described and nectar standing crop production measured. Days to flowering were defined as the number of days from germination to when 10% of the plant populations flowered. Nectar standing crop on the hand was defined as the amount of nectar available to pollinators at a single point in time. It was measured from randomly selected flowers using micro-capillary tubes. The length of the nectar in the capillary tube was measured using a digital vernier calliper and recorded. Nectar volume was calculated as indicated in Cruden and Hermann (1983). For the measurement of solute concentration (percentage sucrose equivalents on a mass basis), the nectar was deposited on the low-volume field hand held refractometer (0–50%, Bellingham and Stanley, Norcross, Georgia, USA) prism and the percent sucrose concentration recorded.

Two flower types, staminate and pistillate flowers were observed on *Cleome gynandra*, therefore, it was necessary to find out the differences in morphologies and their role in pollination of this crop. Further descriptions were made on the length of androgynophore, gynophores, and stamens in both flowers types. Fresh flowers were collected and the length of the androgynophore,

gynophores and stamens were measured using a digital calliper then rounded off to 0.01 mm. *C.* gynandra flowers attracted high number of illegitimate flowers visitors especially honeybees that collected both nectar and pollen. The effect of the illegitimate visitors on nectar volume was determined by simulating illegitimate nectar removal. Randomly selected flowers (n = 24) were guarded from bee visits and labeled. Then, nectar was extracted using 1 μ l micro-capillary tubes and measured from the same flowers every 1 hour beginning 1700 hours to 2000 hours.

Pollination needs of African Indigenous Leafy Vegetables

To find out the pollination needs and the dependence of the various AILVs on pollinator, appropriate pollination treatments were performed and allocated in Complete Random Design (CRD) to the different AILVs flowers. They were carried out depending on the pollination syndrome exhibited by the different AILVs flowers. *Cleome gynandra* exhibited phalaenophily pollination syndromes (nocturnal pollination by moths) while *Crotalaria brevidens* and *Solanum scabrum* displayed melittophily pollination syndrome (pollination by bees).

Due to the different flower morphology and pollination syndrome exhibited by C. gynandra flowers, the following pollination treatments were carried out on pistillate flowers. (1) Hand crosspollination at dusk (HCPD) was performed between 1900 and 2000 hours by dubbing pollen from fully opened anther on the stigma of a different plant, then the anthers on the treated flower were cut off using a pair of scissors and the flower bagged immediately using fine mesh bag to exclude pollinators. (2) Hand cross-pollination at morning (HCPM), was performed between 0600 and 0700 hours by dubbing pollen from fully opened anthers on the stigmas of a different plant, then the anthers on the treated flower were cut off using a pair of scissors and the flower bagged immediately using of 1mm netting to exclude pollinators. (3) Hand self-pollination was carried out at dusk (HSP) stigmas were dusted with pollen from anthers of the same flowers and the pollinated flowers bagged to exclude pollinators. (4) Bagging throughout (BT) (to test the possibility of autogamous pollination) was performed by covering flowers with fine mesh to exclude pollinators, in (5) diurnal pollination (DP) where flowers were opened from 0600 to 1800 daily for access by the diurnal pollinators but bagged from 1800 pm to 0600 hours to exclude nocturnal pollinators, (6) nocturnal pollination (NP) flowers were excluded from visitors from 0600 hours to 1800 and only allowed pollinator visitations from 1830 to 0600. (7) Open pollination (OP - control) where the flowers were tagged and left open both day and night.

Pollination treatments performed for *C. brevidens* and *S. scabrum* were as follows, (1) open pollination (OP - control) with unrestricted visits by the pollinators, (2) bagging throughout (BT) where pollinators were excluded by covering the flowers with fine mesh bags throughout the observation period, (3) hand self-pollination (HSP) where were hand pollinated using pollen from the same flower then bagged using fine mesh material to exclude any further visits, (4) hand cross-pollination (HCP) where flowers were hand pollinated using pollen from the same flower then bagged using fine mesh material and (5) pollen augmentation (PA) flowers were hand cross-pollinated and then left open allowing for further unrestricted visits.

In all the above treatments, bagging materials were removed after fruit set to allow for fruits to develop and mature. Yield parameters, fruit weight and length, number of mature and deformed seeds per fruit and weight of dry mature seeds were recorded. The fresh mature seeds were sun dried under the shade for 7 days to lower the seed moisture level and later dried in the oven at 40°C for 6 hours. They were then measured to get dry seed weight (g).

3.2.3 Dependence of African Indigenous Leafy Vegetables on pollinators for seed set

Dependence of AILVs on pollinators for seed set was determined using established procedures by Morse and Calderone (2001) simply expressed as:

$$D = \frac{Yub - Yb}{Yub}$$

Where:

 $D \Rightarrow$ ratio that ranges from 0 to 1 denoting the contribution of pollinators to crop yield,

 $Yub \Rightarrow$ are yields of the crop obtained from plots that are freely accessed by insect pollinator,

 $Yb \Rightarrow$ are yields of the crop obtained from plots that are not accessed by insect pollinators.

Zero (0) value implies there is no (negligible) additional yield gain from pollinated flowers compared to the yield obtained from un-pollinated flowers, and hence pollinators may not be required while one (1) means that without pollinators the crop cannot reproduce.

3.2.4 Effects of pollination treatments on seed germination rate and total reproductive success (%) of African Indigenous Leafy Vegetables

Seeds from the same treatment were pooled together. Fifty (50) seeds were randomly selected to form a single replicate. Each pollination treatment was replicated 4 times. The seeds were placed on top of blotting papers in Petri-dishes then moistened with distilled water and kept under room temperature. Water was added as needed and the number of germinated seeds counted for a period of 10 days starting from the next day after treatment initiation. The percent germination was calculated as the total number of germinated seeds relative to total number of seeds initiated. Total reproductive success (%) was calculated as the product of the fruit set (%), seed rate (%) and the germination rate (%) per treatment. Since the number of ovules per flower that would set seed was not determined for the AILVs, it was assumed that, the pollination treatment with the highest number of mature seeds per flower was the maximum possible seed set. Seed rate was then calculated as the highest number of mature seeds set in this pollination treatment in relation to the other treatments. Total reproductive success indicates the total viable seeds per flower by a given pollination treatment.

3.2.5 Influence of natural habitat on the seed set and quality of *Cleome* gynandra

To evaluate the role of the Kakamega forest on the hawkmoth pollination and the resultant seed yield and quality, a line transect at 0, 0.5, 1.0, 1.5, 2.0, 4.0, 8.0 km from Kakamega forest edge was developed. At each site, *C. gynandra* was maintained as 30 potted plants. These were maintained throughout the period following recommended agronomic practices. At flowering, 20 flowers were randomly tagged. At maturation, the siliques were harvested then the length and weight measured. The total number of mature and deformed seeds as well as seed weight per silique was recorded.
3.2.6 Data analysis

All data were analysed using the General Linear Models procedure (GLM) (SPSS version 19). One-way Analysis of Variance (ANOVA) was done to compare differences between the pollination treatments at 95% significance level. The means were separated using the Student-Newman-Keuls (S-N-K). Pearson correlation test was performed at 95% significance level, to find out the effect of time of nectar removal by illegitimate floral visitors and the amount secreted in *Cleome gynandar* flowers. In addition, simple linear regression analysis was conducted at 95% significance level to investigate the influence of the distance from the natural habitat (forest) on the seed set for *C. gynandra*. The results were considered significant when $p \le 0.05$, highly significant if $p \le 0.001$, marginally significant if $0.10 \ge p \le 0.05$.

3.3 Results

3.3.1 Flower morphology, nectar standing crop, pollination needs and seed quality of spider plant (*Cleome gynandra* L.)

Flower morphology and nectar standing crop of Cleome gynandra

Flowering started on the 11th May 2010; 30 days from seedling emergence. *C. gynandra* is andromonoecious with both hermaphrodite and staminate flowers within one individual inflorescence. Both flower types provide nectar and pollen as rewards and are only opened for one day. Hermaphrodite flowers have six long stamens and one long gynoecium (functional) (see Figure 3.1a) while staminate flowers have six long stamens with a rudimentary gynoecium that aborted the next day after flower opening (see Figure 3.1b). In both cases the anthers are held away from the sticky gynoecium. Though not measured in this study, by dusk 1845 hours, the flowers are luminous white with a lot of nectar at the petal bases (Figure 3.1c) with conspicuous amount of orange coloured dehisced pollen observed on the anthers (Figure 3.1d). The flowers gave off a faint sweet fragrance by dusk.

ANOVA result on the floral structures for hermaphrodite flowers as compared with the staminate flowers showed significant differences in the length of androgynophore ($F_{1, 41} = 211.733$, p = 0.000), length of gynophore ($F_{1, 41} = 396.596$, p = 0.000), but not stamen length ($F_{1, 18} = 1.006$, p =

0.329) (Table 3.2). Nectar secretion started by 1700 hours. Nectar standing crop was significantly different across the day ($F_{4, 457} = 223.486$, p = 0.000). Nectar amounts increased from 1.2 µl at 1700 hours to about 18.6 µl by 2000. Nectar concentration was not significantly different across the evening ($F_{4, 445} = 0.753$, p = 0.556) but remained constant at mean percent nectar concentration of (± s.e) 12.8% ± 0.57 (n = 450) (Figure 3.2). In the experiment to mimic nectar removal by either the legitimate pollinator or illegitimate (the nectar thieves), it was observed that more nectar was secreted taking approximately 30 minutes to form a droplet at the base of the flower petals. A 2-tailed Pearson's correlation test showed a strong positive correlation between the amount of nectar secreted within one hour and the time of the removal (r = 0.771, n = 84, p = 0.01). More nectar was secreted between 2000 and 2100 hours compared to between 1700 and 1800 hours. Some nectar drops were observed on the flowers early the next day morning.



Figure 3.1: (a) Hermaphrodite flowers (with six long stamens and a long functional gynoecium); (b) arrow showing staminate flowers with long stamens and rudimentary abortive gynoecium (c) arrow showing nectar droplet at the petal bases of *C. gynandra* flowers at 1657 hours; (d) appearance of *C. gynandra* at night with orange pollen on the anthers and luminous white flower petals.

Table 3.2: Floral structures, length of hermaphrodite and staminate flowers of *Cleome gynandra* in Kakamega forest farmlands, 2010

Floral structures	Hermaphrodite (mm) (mean length ±s.e)	Staminate (mm) (mean length \pm s.e)
Androgynophore	$36.6 \pm 0.88 \ (n = 22)$	$18.4 \pm 0.90 \ (n = 21)^a$
Gynophore	$23.2 \pm 0.68 \ (n = 22)$	$4.0 \pm 0.69 (n = 21)^{a}$
Stamen	$28.1 \pm 1.41 \ (n = 10)$	$26.1 \pm 1.41 \ (n = 10)$



Figure 3.2: Mean nectar standing crop volume (μ l) (± s.e) and % sucrose concentration (± s.e) measured at different times in the evening on *Cleome gynandra* flowers.

Pollination needs of Cleome gynandra and dependence of pollinator for seed set

The results on fruit set indicated that *Cleome gynandra* is self compatible but nonautogamous. Flowers bagged to exclude pollinators aborted setting no further seeds. Dependence on pollinators for seed set was high (0.99). Significant differences were observed for silique (fruit) mean length ($F_{5, 170} = 5.010$, p = 0.000), weight ($F_{5, 170} = 11.634$, p = 0.000) and the mean number of mature seeds ($F_{5, 169} = 22.627$, p = 0.000). Hand cross-pollination at dusk had the highest mean silique length, weight and mean number of mature seeds per silique. Hand cross-pollination at dusk increased the number of mature seed per silique by 82% compared to the control (unrestricted floral visitor access). The number of deformed seeds per silique was significantly different ($F_{5, 156} = 2.829$, p = 0.018) and high in hand cross-pollination treatments. Mature seed weight per silique was significantly different ($F_{5, 169} = 18.881$, p = 0.000) with higher seed weights per silique recorded in hand cross-pollination at dusk and in the morning treatments (Table 3.3).

Table 3.3: Percent fruit set, mean fruit length, number of mature seeds, deformed, and weight per fruit (silique) for pollination treatments of *Cleome gynandra* L. in Kakamega forest farmlands, 2010

Pollination	Fruit	Mean weight of	Mean length of	Mean no. of mature	Mean no. of	Mean weight of
treatments	set	silique (g) \pm s.e	silique (cm) \pm s.e	seeds per silique ± s.e	deformed seeds	seeds per silique
	(%)				per silique ± s.e	$(g) \pm s.e$
DP	75.0	$0.3 \pm 0.03^{b}(32)$	8.4 ± 0.53^{b} (32)	$73.8 \pm 11.4^{\text{cd}}$ (32)	12.6 ± 4.84^{ab} (29)	0.1 ± 0.02^{bc} (32)
NP	89.2	$0.5 \pm 0.05^{b} (37)$	$9.5 \pm 0.45^{b} (37)$	$121.2 \pm 10.63^{b} (37)$	$7.4 \pm 1.68^{ab}(37)$	$0.2 \pm 0.02^{b} (37)$
OP	88.9	$0.5 \pm 0.05^{b} (27)$	$9.1 \pm 0.50^{b} (27)$	$120.4 \pm 12.45^{b}(27)$	$5.7 \pm 1.8^{ab} (27)$	$0.2 \pm 0.02^{b} (27)$
HCPM	85.0	$0.5 \pm 0.06^{b} (27)$	$9.0 \pm 0.75^{b}(27)$	109.9 ± 12.68 ^b (26)	$19.9 \pm 4.83^{a}(22)$	$0.2 \pm 0.02^{b} (26)$
HCPD	93.8	$0.8 \pm 0.06^{a} (32)$	$11.4 \pm 0.50^{a}(32)$	$219.3 \pm 11.43^{a}(32)$	$16.7 \pm 4.64^{ab} (32)$	$0.3 \pm 0.01^{a}(32)$
HSP	57.1	$0.3 \pm 0.05^{b}(21)$	8.0 ± 0.36^{d} (21)	$56.7 \pm 14.11^{d}(21)$	$2.4 \pm 0.55^{b}(15)$	$0.08 \pm 0.01^{\rm c} (21)$

Means with same superscript letter within a column are not significantly different at 95% significance level using S-N-K. NP = nocturnal pollination, DP = diurnal pollination, OP = open pollination/Control, HCPM = hand cross-pollination in the morning, HCPD = hand cross-pollination at dusk, HSP = hand self-pollination. Bagging throughtout (BT) was excluded from this analysis. Numbers in parentheses represents N value.

Effect of pollination on seed germination rate and reproductive success (%) of Cleome gynandra

ANOVA results indicated significant differences in rate of germination of seeds from different pollination treatments ($F_{5, 16}$ = 3.652, p = 0.021). Hand self-pollination had the lowest rate of germination. Bagging treatment was excluded from germination tests due to insufficient number of seeds. Higher reproductive success was realized in hand cross-pollination at dusk compared to the hand cross-pollination in the morning and open pollination (control). Hand self-pollination recorded the least reproductive success indicating preference for out-crossing in this species (Figure 3.3).



Figure 3.3: Mean seed germination rate (\pm s.e) and total reproductive success (%) of *Cleome gynandra* with different pollination treatments; NP = nocturnal pollination, DP = diurnal pollination, OP = open pollination/Control, HCPM = hand cross-pollination in the morning, HCPD = hand cross-pollination at dusk, HSP=hand self-pollination. Bagging throughout (BT) was excluded from this analysis. Bars are standard error of the mean.

Effect of distance from the forest on seed set of Cleome gynandra

Distance from the forest edge did not affect the length ($R^2 = 0.032$, p = 0.262, $F_{6, 242} = 1.292$) and weight of siliques ($R^2 = 0.043$, p = 0.106, $F_{6, 242} = 1.769$), number of deformed seeds ($R^2 = 0.033$, p = 0.257, $F_{6, 239} = 1.302$) or weight of mature seeds ($R^2 = 0.036$, p = 0.192, $F_{6, 241} = 1.462$), but had a marginally significant effect on the number of mature seeds per silique ($R^2 = 0.052$, p = 0.055, $F_{6, 237} = 2.096$). Although, as the distance from the forest increased, the number of mature seeds decreased per silique however the effect was very small, $R^2 = 0.052$, (Figure 3.4).



Figure 3.4: Effect of distance from the forest edge on the number of mature seed per silique of *Cleome gynandra* in 2010. Transect points: 0, 0.5, 1.0, 1.5, 2.0, 4.0, 8.0 km from Kakamega forest edge.

3.3.2 Flower morphology, pollination needs and seed quality of slender leaf *Crotalaria brevidens* Benth

Flower morphology and nectar standing crop production

Flowering started 60 days from seedling emergence. The flowers opened for one day and by the next day, they wither off. The floral morphology fits the "keel blossom" with the wing-keel complex (Westerkamp, 1997). They are yellow in colour with a flag (vexillum) petal, two wings on each side and two-fused keel petals (Figure 3.5). The nectar is secreted at the base of the ovary and accumulates at the nectar chamber within the staminal tube, which is closed from the sides by the two wing petals and reinforced from the top by the vexillum (Etcheverry *et al.*, 2003). The vexillum restricts access to the nectar to only specific floral visitors. Seemingly, only large long-tongued bees are able to manipulate the flowers to gain access to the nectar and pollen rewards.

The ANOVA results on the nectar standing crop indicated significant variations with time of the day ($F_{5, 54} = 32.743$; p = 0.000). Higher volumes of nectar (mean ± s.e) 0.85 µl ± 0.60 were recorded at 0900 hours compared to 0.47 µl ± 0.60 at 1000 hours, 0.02 µl ± 0.60 at 1300 hours and

thereafter reduced significantly to zero by 1400 hours. There were records of some individual flowers with high nectar amounts in the afternoon.



Figure 3.5: Crotalaria brevidens flower morphology.

Pollination needs of Crotalaria brevidens and dependence on pollinators for seed set

The results on percentage fruit set from pollinator exclusion treatment indicated that *C*. *brevidens* relies heavily on pollinator fro fruit and seed set with little capability of spontaneous self-pollination (Table 3.4). Dependence on the pollinators for seed set was high (0.98). In hand self-pollination treatment where flowers were manipulated to mimick legitimate visits by pollinators, percent fruit set improved by 44% compared to the fruit set when pollinators were excluded. Fruit set in flowers that received unrestricted pollinator visits and those that were hand cross-pollinated were comparable. In contrast, pollen augmentation did not result into increased percent fruit set (75%) (Table 3.4). Pollination treatments had highly significant differences for both fruit weight (F_{4, 187} = 16.367, p = 0.000), length (F_{4, 187} = 37.850, p = 0.000), number of mature seed per fruit (F_{4, 187} = 49.406, p = 0.000), number of deformed seed per fruit (F_{4, 186} = 4.891, p = 0.001), and the weight of dry mature seeds (F_{4, 187} = 31.761, p = 0.000). In the pollinator exclusion treatment, fruit weight,

length number of mature seeds and the weight of dry mature seeds were lower than observed for other pollination treatments. Hand cross-pollination resulted into heavier fruits, higher number of mature seeds per fruit and heavier dry seed per fruit compared to supplementary pollination where augmented pollen deposition from the presumed additional pollinator visitation did not result into increased yield parameters. Pollinator visitation resulted into 98% increase in mature number of seeds per fruit compared to pollinator exclusion (Table 3.4).

Table 3.4: Percent fruit set, mean fruit weight, length, number of mature seeds, deformed seeds and weight of dry mature seeds of *Crotalaria brevidens*, in Kakamega forest farmlands, 2009

Treatments/Yield	BT (n = 42)	HCP $(n = 31)$	$\mathrm{HSP}\ (\mathrm{n}=49)$	OP (n = 50)	PA (n = 20)
Fruit set (%)	21.4	96.8	65.3	94.0	74.0
Fruit weight $(g) \pm s.e$	0.2 ± 0.11 ^c	1.5 ± 0.13^{a}	0.8 ± 0.11 ^b	1.2 ± 0.10^{a}	1.1 ± 0.16 ^{ab}
Fruit length (cm) \pm s.e	0.8 ± 0.35 ^c	$6.2 \pm 0.40^{\text{ a}}$	4.0 ± 0.32 ^b	6.0 ± 0.32 ^a	$4.8 \pm 0.50^{\text{ b}}$
No. mature of seeds \pm s.e	1.5 ± 3.81 ^d	70.4 ± 4.44 ^a	25.7 ± 3.53 ^c	60.1 ± 3.5^{a}	41.7 ± 5.53 ^b
No. of deformed seeds/fruit \pm s.e	0.1 ± 0.49 ^b	2.8 ± 0.57 ^a	2.2 ± 0.45 ^a	2.6 ± 0.45 ^a	1.6 ± 0.73^{a}
Dry seed weight $(g) \pm s.e$	-	0.4 ± 0.03 ^a	0.1 ± 0.03 ^c	0.3 ± 0.03 ^b	0.3 ± 0.04 ^b

Means with the same superscript letter within a row are not significantly different at 95% significance level using S-N-K for the pollination treatments; BT = bagging throughout, HCP = hand cross-pollination, HSP = hand self-pollination, OP = open pollination/Control, PA = pollen augmentation.

Effects of pollination treatments on germination and total reproductive success (%) of Crotalaria brevidens

Germination started on the third day from experiment initiation date. Lack of pollinators reduced significantly the rate of germination ($F_{4, 15} = 17.481$, p = 0.001). Low mean rate of germination of 30% was recorded from bagging treatments where pollinators were excluded. Higher total reproductive success was recorded for the hand cross-pollination (74%) while hand self-pollination recorded (13.8%), bagging treatment recorded the least reproductive success (0.1%), and was therefore, not included in the graph (Figure 3.6).



Figure 3.6: Mean seed germination rate (%) (\pm s.e) and total reproductive success (%) of *Crotalaria brevidens* seeds of different pollination treatments; BT = bagging throughout, HCP = hand cross-pollination, HSP = hand self-pollination, OP = open pollination/Control, PA = pollen augmentation. Reproductive success for BT treatment (0.14%) excluded from graph.

3.3.3 Flower morphology, pollination needs and seed quality of *Solanum* scabrum

Flowering started 60 days from seedling emergence. The flowers are white in color, small and adapted for self-pollination. The likely reward to pollinators was pollen. Fruit set was low in the augmented pollination but remained high in the other treatments (Table 3.5). Pollination treatment results indicated significant differences among the treatments, mean fruit weight ($F_{3, 182} = 23.365$, p = 0.000), mean longitudinal diameter ($F_{3, 182} = 11.482$, p = 0.000), and mean number of seeds per fruit ($F_{3, 181} = 22.274$, p = 0.000). Dependence of *S. scabrum* on pollinators for seed set was low (0.1), and therefore it can set seeds without any further requirement of pollinator visits.

Effects of pollination treatments on germination rate (%) and total reproductive success (%) of Solanum scabrum

Germination started on the third day from the experiment initiation. ANOVA results showed that germination rate was not significantly different amongst the pollination treatments ($F_{3, 12} = 1.270$, p = 0.329), mean rate of germination of 77.9 ± 4.59, n = 16. However, the total reproductive success was high in open pollination (92%) compared to pollen augmentation (40%), bagging (71%) and hand self-pollination (44%).

Table 3.5: Percent fruit set, mean weight, diameter and number of mature seeds of *Solanum scabrum* in Kakamega forest farmlands, 2009

Pollination treatment/yield	OP $(n = 48)$	BT $(n = 47)$	PA(n = 47)	HSP $(n = 44)$
parameter				
Fruit set $(\%) \pm s.e$	100	100	79.0	91.0
Fruit weight $(g) \pm s.e$	1.5 ± 0.05^{a}	1.3 ± 0.05 ^b	0.9 ± 0.05 ^c	1.0 ± 0.05 ^c
Fruit diameter (cm) \pm s.e	2.1 ± 0.08 ^a	2.0 ± 0.08 ^a	1.5 ± 0.08 ^b	1.7 ± 0.08 ^b
No. mature of seeds/fruit \pm s.e	79.3 ± 2.89 ^a	74.3 ± 2.89^{a}	50.6 ± 2.93 ^b	57.5 ± 2.99 ^b

Means with the same superscript letter within a row are not significantly different at 95% significance level using S-N-K for the pollination treatments; OP = open pollination/Control, BT = bagging throughout, PA = pollen augmentation, HSP = hand self-pollination (HSP).

3.4 Discussion

Pollination needs and influence of pollinators on seed quality of Cleome gynandra L.

Flower morphology, nocturnal anthesis and nectar concentration displayed by *C. gynandra* are attributes for phalaenophily pollination syndrome (nocturnally moth-pollinated flowers). Observed luminous white flowers, low nectar concentration, and faint sweet fragrance by dusk indicates that *C. gynandra* is adapted to nocturnal pollination, most likely by hawkmoths. Similar records of low nectar concentrations, nocturnal anthesis and receptivity have been reported on other *Cleome* species. *Cleome spinosa* in Brazil (Machado *et al.*, 2006), *C. serrulata* and *C. lutea* in USA (Cane, 2008). Low percent nectar concentrations of 12-13% recorded were comparable to those reported by Machado *et al.* (2006) on *C. spinosa* in Brazil. Such low nectar concentrations have been

known to allow for quick nectar uptake by moth pollinators (Baker and Baker, 1982; Machado et al., 2006; Martins and Johnson, 2007). In this case, therefore, the most probable legitimate pollinator would be the hawkmoths. They were observed visiting the flowers for nectar rewards. Apparently, hawkmoths require conspicuous amounts of nectar to compensate for the energy spent during hovering while foraging (Faegri and van der Pijl, 1979). The continual nectar secretion in doses compensates for the lost nectar either due to uptake by the legitimate of illegitimate floral visitor. Further, compensation for nectar is achieved by the aggregations of several opened individual flowers within an inflorescence. Andromonoecy, presence of both staminate and hermaphrodite flowers in one individual observed in this study have also been recorded on other related species for example, C. spinosa (Machado et al., 2006), C. serrulata, and C. lutea (Cane, 2008). It is believed that staminate flowers may increase both nectar availability and pollen flow and thus increase the frequency of pollinator visit, high pollen load on the pollinator's body and consequently more pollen deposits on the stigmatic surface of the flower. All these characteristics may act to reduce the number of flowers visited per inflorescence and encourage more between plants movements, thus achieving high out-crossing and subsequently, seed set. Future measurements of the total nectar produced per flower or inflorescence per night should be encouraged. C. gynandra could be a model plant for studies that are aimed at understanding mechanisms and the energetics of nectar and pollen compensation by plants and how they affect the reproductive success of the plant.

Yield results showed that *C. gynandra* is self-compatible but non-autogamous and therefore relies heavily on pollen vectors. This was shown by the poor seed set in the pollination treatments where pollinators were excluded. Higher fruit set in the hand cross-pollination in the morning could hint towards an extended period of anthesis until early morning that would probably enhance chances of pollination. Total reproductive success improved in hand cross-pollination as result of increased seed set per silique. When hand cross-pollination was performed in the morning, the reproductive success was reduced to (37%). Although fruit set was enhanced, pollen viability may have reduced by the next morning of flower opening. Poor reproductive success in the control and nocturnal pollination could be due to lower incidence of pollination by hawkmoths given such a high stand of *C. gynandra* flowers. Perhaps under natural conditions, poor reproductive success should be expected. However, it was still unclear whether there is strong evidence that lack of sufficient pollination would result into low seed germination rate. Inherent seed dormancy has been associated with the poor germination in *C. gynandra* and seed dormancy breaking treatments, among them hot water treatment, have been recommended (Abukutsa-Onyango, 2007; Ochuodho and Modi, 2005;

Ekpong, 2009). For quality seed control purposes, more urgent research is needed on the acceptable, minimum seed germination rate expected when optimum seed production requirements are given.

Location of seed production site from a natural forest habit is unlikely to influence the seed set of C. gynandra. Although the influence on the distance from the forest edge on the number of mature seed per silique was low, it is possible that plants that were near the edge may have received higher visitation rates by hawkmoths, however this was not reflected on the seed set. Despite the fact that hawkmoths can forage over long distances (Grant, 1983), reductions in moth species richness in distrurbed areas is a concern and can lead to poor pollination, more so for hawkmoth pollinated crops (Beck et al., 2002; Martins and Johnson, 2009). This is however, dissimilar to the results reported by Axmacher et al. (2008) in a study of diversity patterns of moths in Kakamega forest. They found low sphingidae population in the forest and higher populations in the farmland area. Factors such as the high diversity of flowering plants of the farm hedgerows, and the fact that hawkmoths can move over long distances in search of nectar could be attributed to such differences. The rarity of the hawkmoths coupled with the increasing cultivation of C. gynandra in reasonably big farms for seed production may make it impractical to manage them for pollination. Nevertheless, habitat changes and, specifically, forest fragmentation could cause disruptions in the hawkmoth plant-pollination mutualism. Emerging uses of C. gynandra such as its potential as poultry feed, seed oil, and for pest control, highlights a clear need to improve seed set and quality.

Pollination needs and influence of pollinators on seed quality of Crotalaria brevidens

The results indicated that *Crotalaria brevidens* L. has floral morphological adaptations towards insect pollination and offers both pollen and nectar as rewards to floral visitors. The flower structure conforms to the wing-keel complex as described by Westerkamp (1997). Pollen is secondarily presented in response to pressure exerted by the bees on the wings and keel. Similar observations have been described on *Crotalaria stipularia* (Etcheverry, 2001) and *C. micans* by Etcheverry (2003). The diurnal decline of nectar standing crop was possibly due to both nectar secretion rates and the uptake rate by floral visitors. Nectar concentration may have been constant due to the structural protective features such as the presence staminal tube and the vexillum. Such structures been indicated to protect the nectar from environmental parameters such as evaporation, rain and mist as well as to ensure that nectar is only accessible to pollinators with specialised

mouthparts (Pacini and Nepi, 2007). As concentration of the nectar reduces, floral visitors' energy intakes are limited by the low energy content. It would then be expected that, floral visitors confronted with low nectar concentration would visit more flowers to satisfy their energy requirements. Time of the day did not have an effect on the number of flowers visited per bout and the duration of such visits. The observed reduction in the standing crop volumes as the day progressed could be as a result of foragers' intake of the same. Reduced nectar amounts may influence the behaviour of floral visitors and the resultant pollen movement. For instance, during the morning hours when nectar amounts are higher, it is likely that floral visitors' movements would be limited to flowers within an individual plant thereby transferring more self-pollen. On the other hand, as the nectar amounts reduced, foragers who encounter flowers with less nectar would move to far away flowers. It is possible that more cross-pollen could have been deposited by floral visitors later in the day. In addition, the presence of few flowers with higher nectar volume than most of the flowers in the afternoon could have encouraged repeated visits amidst reducing nectar volumes and kept floral visitors active throughout the day thus ensuring that all the opened flowers are visited and pollinated. Southwick (1982) has already described this phenomenon as "lucky hits."

The low percentage fruit set from pollinator exclusion treatment indicates C. brevidens relies largely on pollinators for fruit and seed set and has little capability of spontaneous self-pollination when pollinators are excluded. The dependence on pollinators was high (0.98) indicating that the seed set would be greatly reduced in this species in the absence of pollinators. Similar results have been reported by Free (1993) for the genus Crotalaria. Additional out-cross pollen in the pollenaugmented treatment did not result into any beneficial effect on the fruit set, yield parameters, germination rate and total reproductive success. This suggests that under conditions with adequate pollinator visitations, fruit and seed set are not pollen limited and supplemental pollen may lead to stigma clogging thereby reducing the fruit and seed set. In the pollinator exclusion treatment, both fruit weight and length, number of mature seeds, weight of dry mature seeds and germination rate were low when compared to other pollination treatments. Low germination rates (40%) have been recorded from spontaneous self-pollinated seeds of C. retusa (Jacobi et al., 2005). In the current study, germination rates above 70% were recorded for hand cross-pollination and open pollination with unrestricted pollinator visits. Higher reproductive success were also recorded for hand crosspollination, a clear indication that appropriate vectors are essential to aid in the cross-pollen movement, and prevention of inbreeding depression. Spontaneous self-pollination induces inbreeding depression resulting in reduced germination and plants growth vigour. Further studies may be required to gain insights on the growth characteristics of seedlings from progenies of self-pollinated seeds.

Pollination needs and influence of pollinators on seed quality of Solanum scabrum

Solanum scabrum is highly self-pollinated and does not rely on pollinator for fruit and seed set. The relative small sized flowers as well as short stigma and longer stamens ensure fruit and seed set even in the absence of pollinators. Any slight agitation of the anthers by wind is probably enough to dislodge pollen on to the stigma. High fruit and seed set were recorded across all the different pollination treatments including bagging treatment where pollinators were excluded. However, lower fruit set in the augmented-pollination treatments could be attributed to injuries caused to the stigma during treatment administrations. Seed germination remained high in all the pollination treatments. Nonetheless, out-crossing as indicated in the open pollination with unrestricted pollinator visits improved the reproductive success of this vegetable and would be desireable.

3.5 Conclusions

Generally, the results in this study highlight the importance of pollinators for the increased seed yield and quality of the selected African Indigenous Vegetables. Except for *Solanum scabrum*, pollinators are essential in the seed production of *Cleome gynandra* and *Crotalaria brevidens*. Current results will therefore be useful in programs aimed at developing new varieties through hybridisation. It is further recommended that for increased seed set as well for improved quality, seed production sites of these AILVs should be located near natural habitat such as forest. Natural habitats and diverse hedgerows provide nectar and pollen sources, nesting sites to pollinators and as such, their conservation and management are important for sustainable agricultural production (Kremen *et al.*, 2002; Klein *et al.*, 2003). Surprisingly, the Kenya Seed Company production sites include Thika and Bungoma located in densely populated urban areas and could be almost devoid of pollinators and natural habitats that support them. The efficiency of such sites could possibly be improved by incorporating strips of undisturbed land within the vicinity of the seed fields to act as nesting grounds for the pollinators. More studies should be undertaken in these seed production areas to determine the status of pollinators and the potential effect on seed set and quality. The increasing interest in AILVs

and the fact that seed companies are interested in production of high quality seeds it's therefore important that research is focused on the modes of enhancing pollinator population, for example through habitat rehabilitation, and reduced use of pesticides by embracing an integrated pest management approach for improved pollination service.

4. DIVERSITY, BEHAVIOR AND EFFECTIVENESS OF POLLINATORS OF VEGETABLES CROPS IN KAKAMEGA FOREST FARMLANDS

4.1 Introduction

Most studies on plant-pollinator interactions have always indicated an array of floral visitors that are associated with resultant fruit and seed set. This is so especially for those studies on crop pollination where fruits and seeds are utilised as human food or livestock feed. Depending on the crop's floral morphology, different crops will attract a wide range of flower visitors whose visit may result into fruit set. However, not all visits by a floral visitor guarantee a flower's reproductive success. Understanding which flower visitors are important in the fruit and seed set is thus crucial in crop pollination management strategies to improve crop yield and quality. It is therefore necessary to distinguish among different kinds of visitors and classify them based on their behaviour in relation to methods of pollen or nectar collection. Floral visitors have been classified according to Inouye (1980) and Roubik (1995) as (a) pollinators: whose visit to collect nectar and /or pollen results into pollination and eventual improved fruit/ seed set, (b) thieves: who obtain the reward without damaging the flower, but do not pollinate it due to a mismatch of morphologies, and (c) robbers: that obtain the reward by damaging floral tissues but do not effect pollination pollinators). While some plants, crops included, are usually considered generalised that is their flowers are visited by multiple pollinator species, others may be more specialised as only a subset of floral visitors visiting such flowers will result into fruit and seed set. At times, the most important pollinator can be predicted from the floral morphology, floral visitor behaviour as well as the abundance of the floral visitors in relation to the other recorded visitors. However, the most abundant floral visitor may not at times be the most effective pollinator. Less effective flower visitors may, under certain conditions, become important pollinators and thus may be critical to the reproduction success of the plant/crop species (Waser et al., 1996). It is therefore important to screen amongst the wide range of flower visitors to ascertain the most effective for fruit and seed set if any managed crop pollination is to be undertaken.

The basic approach used to estimate a given pollinator's contribution to pollination involves observing flower visits and evaluating variables such as visitation frequency and visitation duration (Primack and Silander, 1975). On the other hand, indirect measures employed to assess the pollination efficiency of flower visitors have relied on the pollen carried by a pollinator as an

indicator of effectiveness. This approach may however be misleading since pollen on the insect body tells nothing of the quality and the viability of the pollen. Further, relative abundance of the pollinator, visitation rates, and the amount of pollen deposited have been used to enhance this indirect measure (Primack and Silander, 1975). How this measure relates to the eventual seed set is nevertheless still questionable. The importance of floral visitors in the reproduction of a crop and especially where reproductive success is pollen-limited, will ultimately depend on their effectiveness as pollinators. Pollination effectiveness is defined as the total contribution to plant reproduction i.e seed set by a particular pollinator and may vary amongst different pollinators due to behavioural and morphological characteristics (Primack and Silander, 1975; Schemske and Horvitz, 1984). It measures the effect on seed set of a plant population on receiving a single visit by a given species of floral visitor (Spears, 1983). On the other hand, pollinator efficiency is defined as the relative ability of a pollinator to seed set per unit measure e.g time or foraging distance. Effective pollinators are those floral visitors that produce a large number of seeds per single visit, while efficient pollinators are those that produce large number of seeds but spending less time per flower. Availability of such information as the most effective and efficient pollinators for a particular crop, are prerequisites for further studies to identify pollinator-specific nesting requirements, and possibly, draft conservation strategies for sustainable crop pollination.

Although, honeybees, leafcutters and bumblebees are used in managed pollination of crops in the developed countries like United States of America, Canada and Europe (Delaplane and Mayer, 2000), they may not be necessarily effective crop pollinators under the production systems in Kenya. This is so due to spatial differences, pollinator diversity as well as agricultural production systems and crop diversity. Furthermore, importation of pollinators may not be warranted because of risks of possible proliferation of weeds, invasive species, and competition with existing native bee fauna. The establishment of the diversity as well as their effectiveness of pollinators of these crops is therefore essential.

Most small-scale farmers in Kenya practice mixed cropping where more than one crop is grown on the same piece of land. Pollinator requirement for each of these crops may be unique and farmers may likely have poor crop yield due to pollination limitation (Kasina, 2007). Despite this, pollinator management is rarely practised and pollination is generally provided by wild un-managed pollinators (Kasina, 2007; Hagen and Kraemer, 2010). Reasons attributed to this include lack of information on pollinator diversity and effectiveness on seed set for most crops. Gikungu (2006) identified over 200 bee species visiting plants including crops in the farmlands around Kakamega. No study to date has evaluated the effectiveness of these pollinators for future managed crop pollination. This experiment was carried out to identify the pollinator guilds and compare their effectiveness in selected crops grown in the farmlands neighbouring Kakamega forest. This will highlight pollinator species of priority for future breeding, mass culture studies for use in managed pollination in both open field and greenhouse crop production systems.

4.2 Materials and Methods

4.2.1 Study site and test crop planting

The study was undertaken within the farmlands surrounding Kakamega forest. The forest is a tropical rainforest remnant located in Western Kenya (0°17'N, 34°54'E) at an altitude of 1,500 m to 1,700 m above sea level, about 50 km north of Lake Victoria (KIFCON, 1994). Peak rainfall occurs in April-May (long rains) and October-November (short rains) with mean monthly temperatures range from 11°C to 29°C with an average daily temperature of 22°C (Jaetzold and Schmidt, 1982). The experiment was conducted during the long rains season (April-May) in 2009 and 2010. Farmlands neighbouring Kakamega forest are heterogeneous with most farmers growing maize and beans, vegetables and fruits (tomatoes, passion fruit, papayas, pumpkins, and bananas) on the same piece of land. Most of these crops rely on pollinators for fruit set (Kasina, 2007).

Study crops included eggplant (*Solanum melongena* L.: Solanaceae) variety black beauty, soya (*Glycine max* (L.) Merr: Fabaceae) variety Nyala, garden peas (*Pisum sativum* L.: Fababceae), okra (*Hibiscus esculentus* L., Malvaceae) variety lady's finger, karela (*Momordica charantia* L.: Cucurbitaceae), spider plant (*Cleome gynandra* L.: Cleomaceae), slender leaf (*Crotalaria brevidens* Benth: Fabaceae) and broad-leafed African nightshade (*Solanum scabrum* Mill: Solanaceae). Most of these crops are grown in the farmlands of Kakamega forest on a small scale level for house hold consumption. Elsewhere these crops are majorly grown for the urban and the export markets (eggplant, karela, okra) and for oil production (soya beans). Plots of 10 m x 10 m were demarcated and seeds of the crops planted. The seeds were sourced from agricultural outlets. DAP fertilizer was applied at a rate of 100 kg ha⁻¹, each plot received 1 kg of DAP at planting. A staggered planting of two weeks between crops allowed for reduced competition for flower visitors between the flowering crops. The spacing used for all the crops was 60 cm x 90 cm. This spacing was enough to allow for sufficient plant population as well as movement of the data collectors during observation period. Direct planting was done in all the crops, sowing three seeds per hole. At germination, thinning was

done leaving only one plant per hole. Eggplants seeds were first planted in the nursery and then transplanted in the seedbed at three leaf stage at a spacing of 60 cm x 90 cm. Weeding was done every three weeks using hand held hoes. Insect pests and diseases were controlled using appropriate insecticides and fungicides whenever necessary. The sprayings were done late in the evening at the end of the week outside data collection days.

4.2.2 Pollinator diversity and behaviour on selected vegetable crops

Spider plant (Cleome gynandra L.)

Due to the phalaenophilly pollination syndromes displayed by *C. gynandra* flowers (Faegri and van der Pijl, 1979), a thorough investigation was carried out to ascertain its pollinators. Flower visitors were observed for a total of 30 days; from 18th May to 18thJune 2010 between 0700 to 0900 hours early morning, then from 1500 to 1800 hours in the afternoon. Mist nets of 9 m long by 3 m tall were erected across the plot to monitor any possible visitations by bats. Diurnal visitors were carefully observed to determine whether they could touch stigmas and anther and recorded in ten randomly marked *C. gynandra* plants. Based on their behaviour on the flower, nectar and pollen robbers/thieves were noted separately according to Roubik (1995). A total of 42 observations hours were made from 1700 to 2145 hours for two weeks. Using a dim spot light, on hearing the hovering sound made by the hawkmoths during foraging, the foraging behaviour was observed in terms of mode of approach, landing, and whether during foraging the essential reproductive flower structures were contacted. Further, caught hawkmoth individuals were relaxed and their proboscises stretched out, measured to the nearest 0.01 mm using a digital vernier calliper and observed in the laboratory under microscope for *C. gynandra* pollen presence on the head, proboscis or body.

Other test crops

Diurnal observations for pollinator diversity and behavior were made between 0900 to 1600 hours for the other crops, namely, eggplant (*Solanum melongena*), soya (*Glycine max*) variety Nyala, garden peas (*Pisum sativum* L.), okra (*Abelmoschus esculentus*), karela/bitter gourd/ampalaya (*Momordica charantia*), slender leaf (*Crotalaria brevidens* Benth) and broad-leafed African nightshade (*Solanum scabrum* Mill). Observations on floral visitors' diversity and foraging

behaviour were made five days a week between 0900 to 1600 hours for approximately six weeks. Legitimate visits by floral visitor were defined as those visits that resulted in successful exposure of the reproductive structures. Total number of foragers was noted at every one (1) hour interval in the whole plot. Careful observations were made while walking along the crop rows on approaching/landing foraging insects. Without disturbance, the forager was followed and records made on the number of visited flowers using a hand held tally counter. The total time the flower visitor took on the plot was also recorded using a stop watch which was stopped as soon as the forager flew away from the plot. Direct attempts were made to identify the insect species visiting the flowers, but foraging species identification was always difficult in the field, therefore, voucher specimens were collected, identified and deposited at the Invertebrate Zoology Laboratories (IZL) of the National Museums of Kenya, Nairobi (NMK). Percent relative pollinator frequency was calculated as the total number of observed cases of a particular pollinator species expressed as a percentage of the total pollinator species observed during the entire pollinator diversity observation period.

4.2.3 Pollinator effectiveness (PE*i*) and efficiency (PE*t*)

Pollinator effectiveness (PE*i*) was defined as the relative contribution to seed set following a single visit by a given pollinator. It indicates the relative influence of the pollinator to the plant reproductive success. Pollination treatments included: (1) no insect pollination (BT), (2) open pollination for unrestricted pollinator access (OP) and (3) single visits from the various pollinators (SV). In single visit, previously bagged flower was exposed to the a given pollinator until it received only a single visit then bagged and tagged with the corresponding name of the particular pollinator visitor. Efforts were made to expose as many flowers to single visits as possible. At harvesting, the number of seeds per fruit for each of the treatments was counted and recorded. Pollinator effectiveness (PE*i*) was calculated as defined by Spears (1983);

$$PEi = \frac{Pi - Z}{U - Z}$$

where:

- P*i* => is the mean number of mature seed set per pod by flowers receiving single visit of a pollinator species *i*,
- Z => is an estimate of the mean number of mature seed set per pod by flowers where pollinators were excluded, and
- U => is an estimate of the mean number of mature seed set per pod by flowers in open pollination receiving unrestricted pollinator visits.

Minimum pollinator visits required for seed set in each crop species was calculated as the inverse of PEi (PEi^{-1}).

Pollination efficiency (PE*t*) was defined as the mean number of seeds set by a single visit per unit time spent per flower during foraging by the pollinator. It was calculated as the mean number of seeds set per single visit as ratio of the total time spent (seconds) per flower per a given pollinator species.

4.2.4 Data analysis

All the data were analysed using the General Linear Models procedure (GLM) (SPSS, version 19) and Analysis of Variance (ANOVA) generated. Significant differences between the pollination treatments were performed at 95% significance level and respective means compared using the Student-Newman-Keuls (S-N-K). The results were considered significant when $p \le 0.05$, highly significant if $p \le 0.001$, marginally significant if $0.10 \ge p \le 0.05$).

4.3 Results

4.3.1 Spider plant (*Cleome gynandra* L.) pollinator diversity, behavior, effectiveness and efficiency

Both diurnal and nocturnal floral visitors were observed visiting C. gynandra. Diurnal flower visitors included Lasioglossum sp. (Halictidae), Apis mellifera, Amegilla sp. (Apidae), and syrphid fly (Syrphididae: Diptera). The latter were considered nectar thieves, while Apis mellifera, Lasioglossum sp. and Amegilla sp. were nectar and pollen thieves and were inconsequencial for the reproductive success of the plant. Visits by diurnal flower visitors were rarely observed outside 0700 to 0900 hours in the morning and 1500 to 1800 hours in the evening. Nocturnal visitors were seven hawkmoth species (Table 4.1; Figure 4.1). Their visits were considered legitimate and coincident with the nocturnal floral anthesis. They collected nectar with their considerably long proboscis (Figure 4.1). ANOVA results indicated significant differences in proboscis length among the hawkmoth species ($F_{6, 50} = 12.192$, p = 0.000). Hippotion species and Xathopan morgani had the shortest proboscis compared to Agrius convolvuli. Over 20% of the collected samples had pollen on their body parts with 60% of observed pollen presence occurring on the proboscises compared to 40% on the head region. Short tongued hawkmoths species Hippotion eson, H. osiris and Nephele *aequivalens* had the majority of cases of C. gynandra pollen presence and were therefore considered pollinators. Though frequent visitors, A. convolvuli were considered nectar thieves and not pollinators as their long proboscides had pollen on only two (2) out of the 36 processed samples (Table 4.1).

Table 4.1: Nocturnal hawkmoth diversity, proboscis length and *Cleome gynandra* pollen presence,2010

Species name	No. of individuals caught	Mean length of proboscis (mm ± s.e)	% of individuals bearing pollen
Agrius convolvuli L.	41	$102.3 \pm 4.08 (36)$	5.6
Coelonia fulvinotata	4	89.9*	0
Hippotion osiris	7	58.1 ± 12.24 (4)	50.0
Hippotion eson	8	45.4 ± 9.25 (7)	20.0
Hippotion balsiminae	3	26.7*	0
Nephele aequivalens	6	59.0 ± 10.0 (6)	33.3
Xanthopan morgani	2	68.0 ± 17.33 (2)	50.0 ^a

**Coelonia fulvinotata* and *Hippotion balsiminae* not included in the ANOVA (n = 1), ^a*Xanthopan morgani* n = 2.



Figure 4.1: Agrius convolvuli with long outstretched proboscis (mean length 102.3 mm \pm 4.08, n = 36), foraging for nectar on *Cleome gynandra* at 1902 hours in the farmlands of Kakamega forest.

4.3.2 Slender leaf (*Crotalaria brevidens* L.) pollinator diversity, behavior, effectiveness and efficiency

Seventeen (17) bee species from two families were recorded visiting *C. brevidens* flowers in the farmlands neighbouring Kakamega forest (Table 4.2, Figure 4.2). These were, Megachilidae (*Megachile bituberculata, M. rufipes, M. dariensis, M. ithanoptera, Megachile* sp. 1-6), Apidae (*Xylocopa calens, X. inconstans, X. nigrita, X. flavorufa, Apis mellifera, Ceratina* sp. and *Thyreus* sp.). *Apis mellifera* and *Ceratina* sp. visiting slender leaf were categorized as nectar thieves as their foraging behaviour was unable to manipulate the flower in a manner to release the reproductive parts and only visited already opened flowers but still they were unable to reach the nectar chamber. Seemingly, only large long-tongued bees were able to manipulate the flowers to access both nectar and pollen rewards. During foraging, the large bees alighted on the petal and while trying to access the nectar, the wing flowers are pushed outwards, the vexillum upwards and the keel petal pushed down. As they simultaneously put more pressure on the keel petals, vexillum petals are pushed further upwards. They then insert their proboscis through the opening on the upper side of stamina tube to access the nectar chamber. Meanwhile, the pollen present in the anther tubes of the stamens

and the stigma are pushed outwards and comes into contact with the ventral side of the forager. At the end of visit, all the floral parts return to their original position except the flag leaf that remains pushed up. On visiting another flower, the process is repeated and as the stigma comes into contact with the pollen previously deposited on the ventral side of the forager thus favouring crosspollination and to some extent self-pollination. This mechanism can be worked several times on the day of the flower opening. The next day, the flowers start to wither off and foragers avoid them.



Figure 4.2: (a) and (b) leaf cutter bees (*Megachile* sp.); (c) and (d) carpenter bees (*Xylocpa calens Xylocopa inconstans* respectively) foraging for nectar from flowers of *Crotalaria brevidens*.

Megachile and *Xylocopa* spp. were considered legitimate floral visitors due to their frequent visits and relatively large sizes that enabled them to manipulate and expose the flowers' reproductive parts. They were observed to make more than one pressing on the keel as they probed for more nectar thus pushing the stigma out to and fro for several times. For *Megachile* species, pollen was deposited on the metasomal ventral scopa fitting very well with the pollination requirement for these flowers. *Xylocopa* spp. lacks the specialised metasomal ventral scopa but had pollen grains deposited laterally on the first segment of the abdominal segment. *Megachile* spp. were observed to visit partially

opened flowers during the afternoon hours. The floral visitors species were significantly different (F_4 , $_{678}$ = 2.493, p = 0.042) in the mean number of flowers visited in a foraging bout. *Megachile* spp. and Xylocopa spp. visited more flowers per foraging bout compared to other floral visitors (Table 4.2). Apart from their infrequent visits, number of flowers visited and the duration of time on them was highly variable amongst Apis mellifera, Thyreus sp., and Ceratina sp. These bee species either flew away as soon as they were unable to access nectar from already opened flowers or stayed exceptionally longer while learning the flower. Time of the day had a marginal effect on the number of flowers visited per foraging bout (p = 0.076) and the foraging bout duration (p = 0.054). Time taken per foraging bout was significantly ($F_{4, 669} = 2.725$, p = 0.029) different for the various floral visitor species. Although the sizes of the observed bee species were not measured, the relatively bigger sized Xylocopa spp. spent more time compared to Megachile spp. during a foraging bout (Table 4.2). They were observed to make more than one pressing on the keel during any visit as they probe deeper and deeper for nectar thus pushing the stigma out to and fro for several times. For Megachile species, pollen was deposited on the metasomal ventral scopa fitting very well with the pollination requirement for these flowers. Xylocopa spp. lacks the specialised metasomal ventral scopa but had pollen grains deposited laterally on the first segment of the abdominal segment.

High fruit set of over 90% were recorded for single visits for both *Megachile* and *Xylocopa* species. ANOVA results indicated significant differences for mean number ($F_{5, 163} = 26.246$, p = 0.000) and weight of dry mature seeds ($F_{5, 163} = 22.844$, p = 0.000). Single visits from *M. bituberculata*, *M. rufipes*, *Xylocopa calens*, and *X. inconstans* were not statistically different from the open pollination (control) treatment but both differed from the bagging (pollinator exclusion) treatment. *Megachile rufipes*, *Xylocopa calens*, *X. inconstans* had higher PE value than *M. bituberculata*. Meanwhile, *M. rufipes* was the most efficient setting 16.2 seeds per second spent on the flowers (see Table 4.3). The reciprocal of the mean Spear's (PE*i*) an estimate of the number of visits a particular pollinator species would have to make to a flower to be able effect the same seed set equivalent to seed set in natural conditions with unrestricted pollinator visits were for *M. bituberculata* (2), *M. rufipes* (1), *X. calens* (1), and *X. inconstans* (1).

Table 4.2: Percent number of bee species observed out of the 1799 observations, behaviour of floral visitors, number of flowers and time spent in a foraging bout to *Crotalaria brevidens* in Kakamega forest farmlands, 2009

Bee species	% Number of observation made	Ability to trip flower	Mean no. of flowers visited in a single foraging bout ± s.e	Mean time spent per foraging bout (seconds) ± s.e
Megachile spp. ^a	90.9	+	11.4 ± 0.51 (525)	38.2 ± 2.59 (525)
Apis mellifera	0.2	-	2.3 ± 6.24 (3)	8.0 ± 30.77 (3)
<i>Xylocopa</i> spp. ^b	8.4	+	13.9 ± 0.96 (157)	53.6 ± 5.1 (157)
Thyreus sp	0.4	-	6.2 ± 3.66 (12)	9.0 ± 18.1 (12)
<i>Ceratina</i> sp	0.1	-	8.5 ± 7.64 (2)	9.5 ± 37.71 (2)
Social bees	0.2	-	-	-
Solitary bees	99.8	+	-	-

Solitary bees were all bees except *Apis mellifera* (social bees). ^aNine *Megachile* spp., ^b four *Xylocopa* spp. Able to trip flower (+), unable to trip flower (-). Number in parentheses represents N value.

Table 4.3: Percent fruit set, mean mature seed and weight of mature seed per pod, pollinator effectiveness (PE*i*) and pollinator efficiency (PE*t*) of different pollinator species visiting *Crotalaria brevidens* in Kakamega forest farmlands, 2009

Bee species /yield parameters	Fruit set (%)	Mean mature seed number ± s.e	Mean dry mature seed weight \pm s.e	PE <i>i</i>	PEt
Bagging $(n = 42)$	21.4	1.5 ± 4.23 °	-	-	-
Open (Control) (n = 50)	94.0	60.1 ± 3.88 ^a	0.3 ± 0.02 ^a	-	-
<i>Megachile bituberculata</i> (n = 14)	92.9	39.8 ± 7.33 ^b	0.2 ± 0.05 ^a	0.65	11.7
<i>M. rufipes</i> $(n = 44)$	95.5	55.0 ± 4.13^{a}	0.3 ± 0.03 ^a	0.91	16.2
<i>Xylocopa calens</i> (n = 11)	90.9	59.5 ± 8.27 ^a	0.2 ± 0.05 ^a	0.99	15.3
<i>X. inconstans</i> $(n = 8)$	75.0	59.5 ± 9.69 ^a	0.3 ± 0.06 ^a	0.99	15.3
<i>X. flavorufa</i> $(n = 1)^*$	100*	45.0*	0.3*	0.74*	

Means followed by the same superscript letter within a column are not significantly different based on Student-Newman-Keuls test at 95% significance level. *X. flavorufa* not included in the ANOVA (n = 1).

4.3.3 Karela (*Momordica charantia* L.) pollinator diversity, behaviour, effectiveness and efficiency

Both staminate (male) and pistillate (female) flowers were observed on *M. charantia*. Ratio of pistillate to staminate flowers was 1:13 in both 2009 and 2010 in the experimental plot. Pistillate flowers are rewardless. Eleven insect species representing two orders (Hymenoptera and Diptera) were recorded (for details see Table 4.4). While honeybees (*Apis mellifera*), *Plebeina hildebrandti* and *Lasioglossum* sp. were frequent visitors, carpenter bees (*Xylocopa* spp.) were considered to be of moderate frequency to the flowers of *M. charantia*. Few observations of *Hypotrigona gribodoi* and only a single record of a muscid fly, *Amegilla* sp. were recorded visiting male flowers only. They were considered to be of negligible reproductive importance and were excluded in the forager behaviour and visitation analysis.

Floral visitors discriminated between the staminate and pistillate flowers. They were most likely to visit more staminate flowers per bout compared to pistillate flowers. In both observation years (2009 and 2010) the proportion of pistillate flowers that received visits by pollinators remained below 1% of the total recorded visits by pollinators. Apis mellifera, Plebeina hildebrandti, X. *flavorufa* and X. *inconstans* visited pistillate flowers, but in very negligible frequencies. It was unlikely that these pollinators made more than one visit to pistillate flowers in any single foraging bout. In most of the cases, a bee landing on a pistillate flower flew away from the study plot without visiting another flower (staminate or pistillate). In fact, pistillate flowers received the recorded visits only by chance (Table 4.4). Only one single visit to pistillate flower was observed by Xylocopa flavorufa in 2009, while occasional single visits were recorded from Lasioglossum sp., Apis mellifera, Plebeina hildebrandti but these visits were brief compared to the duration of the visits by the same pollinator species to the staminate flowers. Irrespective of the pollinator species, pistillate flowers were only visited once within any foraging bout in most cases of the few recorded visits to pistillate flowers, a visitor landing on the flower flew away from the plot almost immediately without visiting any other flowers. But, before flying away from the pistillate flowers, Apis mellifera, Lasioglossum sp. and P. hildebrandti were observed to search for nectar reward from the base of the stigma (Figure 4.3). In both years 2009 and 2010, all bagged flowers (pollinator exclusion) aborted. Fruit set in both years was low for the open pollination treatments, but improved with additional manual pollen deposition on the pistillate flowers (Table 4.5 and 4.6). Hand cross-pollination resulted in better fruit set 60% in 2009 (n = 15) and 44% in 2010 (n = 18) as compared to the open

pollination 38% (n = 24) and 14% (n = 37) respectively. A few single visits by A. mellifera, Lasioglossum sp. and P. hildebrandti were recorded in 2010.

Table 4.4: Floral visitors observed on the flowers of *Momordica charantia* and their proportional visits to staminate and pistillate flowers, in Kakamega forest farmlands, 2009 and 2010

Bee family/Year	Species name	No. of visitations observed	% Relative frequency	Total no. of staminate flowers visited *	Total no. of pistillate flowers visited
2009	_				
Apidae	Apis mellifera	507	46.6	1705	3
	Plebeina hildebrandti	300	27.6	487	3
	Xylocopa calens	62	5.7	239	0
	X. flavorufa	53	4.9	211	1
	X. inconstans	34	3.1	138	0
	X. nigrita	25	2.3	102	0
	Amegilla sp.	1	0.1	2	0
	Hypotrigona gribodoi	1	0.1	1	0
Subtotals		983			
Halictidae	Lasioglossum sp.	110	10.1	123	0
Megachilidae	Megachile sp.	1	0.1	1	0
	Muscid fly	1	0.1	2	0
Totals		1087	100	3011	7
2010					
Apidae	A. mellifera	281	60.8	447	2
-	P. hildebrandti	52	11.3	56	0
	X. calens	4	0.9	17	0
	X. flavorufa	3	0.6	13	0
	X. inconstans	5	1.1	17	0
	X .nigrita	4	0.9	16	0
Subtotals	-	349			
Halictidae	Lasioglossum sp.	113	24.5	117	1
	Totals	462	100	683	3

*Includes also multiple visits to flowers during an observation.

Figure 4.3: (a) Lasioglossum sp.; and (b) Apis mellifera collecting pollen from male flowers of Momordica charantia.

Of the 14 single visits recorded for *A. mellifera*, 86% set fruit and 14% aborted. All the single visits by *P. hildebrandti* (n = 2) and *Lasioglossum* sp. (n = 3) set fruit. ANOVA indicated a highly significant difference among the pollination treatments. Weight of the fruit was significantly different in 2010 ($F_{2, 85} = 7.608$, p = 0.001 in 2010) but only marginally in 2009 ($F_{2, 57} = 2.589$, p = 0.084). Hand cross-pollinated fruits were heavier than other pollination treatments (Table 4.5).

Table 4.5: Mean yield parameters for the different pollination treatment in *Momordica charantia* in Kakamega forest farmlands, 2009

Treatments/Yield parameters	OP (n = 24)	PA (n = 21)	HCP $(n = 15)$
Fruit set (%)	38	52	60
Weight of fruit (g) \pm s.e.	2.9 ± 1.90^{a}	4.3 ± 2.03^{a}	9.7 ± 2.41 ^a
Length of fruit (cm) \pm s.e.	1.7 ± 1.09 ^b	3.6 ± 1.17^{b}	8.3 ± 1.38^{a}
No. of mature seed /fruit ±s.e.	1.0 ± 1.23 ^b	1.7 ± 1.32 ^b	8.5 ± 1.56^{a}
Weight of mature seeds /fruit (g) \pm s.e.	0.1 ± 0.06 ^a	0.1 ± 0.06 ^a	0.06 ± 0.06 ^a

Means followed by the same letter in superscript within arrow are not significantly different Student-Newman-Keuls based on test at 95% significance level. Number in parentheses represents N value, OP = open pollination, PA = pollen augmentation, HCP = Hand cross-pollination.

The length of the fruits were significantly different ($F_{2, 57} = 7.297$, p = 0.002 in 2009 and $F_{2, 85} = 11.793$, p = 0.000 in 2010). Longer fruits were recorded in both hand cross-pollination and in pollen augmentation treatments (Table 3.5 and 3.6). The number of mature seeds per fruit were found to be significantly different between pollination treatments ($F_{2, 57} = 7.891$, p = 0.001 in 2009 and $F_{2, 88} = 7.835$, p = 0.001 in 2010). The weight of mature seeds were not significantly different among pollination treatments in 2009 ($F_{2, 57} = 0.473$, p = 0.625) but was significantly different in 2010 ($F_{2, 85} = 7.264$, p = 0.001). Both hand and supplemental hand cross-pollination resulted in higher fruit weight, length and mature seed numbers compared to natural pollination (Table 4.6).

Fruit weight, length, number of mature seeds and weight of dry seeds were not significantly different (P>0.05) amongst the single visits by *Apis mellifera*, *P. hildebrandti* and *Lasioglossum* sp. Pollination effectiveness was 0.86 for *Apis mellifera*, 0.88 for both *P. hildebrandti* and *Lasioglossum* sp. *Apis mellifera* was the most efficient setting about 1.3 seeds per second, while *P. hildebrandti* and *Lasiglossum*, was 0.2 and 0.1 seeds set per second of visit respectively.

Table 4.6: Mean yield parameters for the different pollination treatment in *Momordica charantia* in Kakamega forest farmlands, 2010

Treatments/Yield parameters	OP (n = 37)	PA (n = 33)	HCP (n = 18)
Fruit set (%)	14	61	44
Weight of fruit $(g) \pm s.e.$	0.6 ± 0.60 ^b	3.9 ± 0.63^{a}	3.0 ± 0.86^{a}
Length of fruit (cm) \pm s.e.	0.8 ± 0.77 ^b	6.2 ± 0.81 ^a	3.8 ± 1.1^{a}
No. of mature seed /fruit \pm s.e.	0.3 ± 0.77 ^b	4.3 ± 0.82^{a}	4.3 ± 1.2^{a}
Weight of mature seeds /fruit $(g) \pm s.e.$	0.04 ± 0.09 ^b	0.6 ± 0.11^{a}	0.4 ± 0.14 ^a

Means followed by the same letter in superscript within a row are not significantly different based on Student-Newman-Keuls test at 95% significance level. Number in parentheses represents N value. OP = open pollination, PA = pollen augmentation, HCP= hand cross-pollination.

4.3.4 Broad-leafed African nightshade (*Solanum scabrum* Mill) pollinator diversity, behaviour and effectiveness

During 180 observation hours, nine bee species were observed visiting *S. scabrum* flowers (Table 4.7). Most of these floral visitors were soil-nesting bees with exception of *Apis mellifera* and *Xylocopa calens*. Bee species *Amegilla langi*, *Nomia* sp. and *X. calens* were able to buzz the flowers releasing a cloud of pollen. *Apis mellifera* on the other hand were observed vigorously striking the anthers with their forelegs and were seen packing the white pollen unto their corbiculae. There were significant differences in the number of flowers visited ($F_{5, 947} = 165.476$, p = 0.000) and time taken ($F_{5, 947} = 104.199$, p = 0.000) during a foraging bout. Although *Apis mellifera* were the most frequent floral visitor, they visited fewer flowers compared to the less frequent *A. langi* and *X. calens*. *A. langi* and *X. calens* buzzed the flowers and were observed to rapidly visit many flowers within a short time during foraging. They were considered the most likely pollinators for this crop (Table 4.7).

The results indicated significant differences among the pollination treatments, mean fruit berries weight ($F_{6, 147} = 7.131$, p = 0.000), mean longitudinal diameter ($F_{6, 147} = 2.956$, p = 0.009) and mean number of seeds per fruit berry ($F_{6, 147} = 5.867$, p = 0.000). In all the treatments, fruit set was 100%. *S. scabrum* can set seeds without any further requirement of pollinator visits. However, fruit berries resulting from single visits from *Apis mellifera* and *Lasioglossum* had the lowest fruit berry weight (1.2 ± 0.05 g and 1.2 ± 0.07 g respectively) while the number of seeds per fruit berry was not different between the single visits from *A. langi* and *X. calens*, bagging (pollinator exclusion) and open (unrestricted pollinator visits) (Table 4.8). Further results from the pollinator effectiveness evaluation of floral visitors to *S. scabrum* revealed that buzz pollinators were the most effective and efficient. *Amegilla langi* and *Xylocopa calens* were effective and efficient when compared to *Nomia* sp., and *Lasioglossum* sp.

Table 4.7: Floral visitors' diversity, frequency of visit and behaviour on flowers of *Solanum scabrum* in Kakamega forest farmlands, 2009

Bee species	Total	% Relative	Mean no. of flowers	Mean time taken
	observations	frequency	visited per bout \pm s.e	per bout (s) \pm s.e
Apis mellifera	368	38.6	16.2 ± 0.43 ^b	49.4 ± 1.37 ^b
Lasioglossum sp.	308	32.3	4.4 ± 0.47 ^c	22.1 ± 1.50 °
Amegilla langi	141	14.8	24.0 ± 0.7 ^a	71.0 ± 2.21 ^a
<i>Nomia</i> sp.	87	9.1	2.8 ± 0.89 ^c	15.7 ± 2.81 ^{c d}
Lipotriches sp.	26	2.7	3.2 ± 1.62 °	$9.8 \pm 5.15^{\text{ d}}$
Xylocopa calens	23	2.3	22.7 ± 1.73^{a}	67.4 ± 5.47 ^a
Thrincostoma torridum*	1	0.1	1*	67*

Means followed by the same letter in superscript within a column are not significantly different based on Student-Newman-Keuls test at 95% significance level. * *Thrincostoma torridum* not included in the ANOVA (n = 1).

Table 4.8: Mean fruit weight, longitudinal diameter, number of seed per fruit and pollination effectiveness of floral visitors on the seed set of *Solanum scabrum* in Kakamega forest farmlands, 2009

Bee species/Yield parameter	Mean weight $(g) \pm s.e$	Longitudinal diameter (cm) ± s.e	No. of seeds per fruit /berry ± s.e	PEi	PEt
OP (n = 48)	1.4 ± 0.03 ^a	2.1 ± 0.03^{a}	80.1 ± 1.58 ^a	-	-
BT (n = 47)	1.3 ± 0.03 bcd	$2.0 \pm 0.03^{\ ab}$	$74.5 \pm 1.60^{\text{ ab}}$	-	-
<i>Apis mellifera</i> (n = 12)	1.2 ± 0.05 ^d	1.8 ± 0.06 ^b	65.3 ± 3.15 bc	-1.6	21.4
<i>Lasioglossum</i> sp. $(n = 6)$	1.2 ± 0.07 ^d	2.0 ± 0.08 ^{ab}	60.5 ± 4.46 ^c	-2.5	12.1
<i>Amegilla langi</i> (n = 16)	1.4 ± 0.04 ^{abc}	2.1 ± 0.05^{a}	76.9 ± 2.73^{a}	0.4	25.6
<i>Xylocopa calens</i> $(n = 14)$	1.4 ± 0.05 ^{ab}	$2.0 \pm 0.05^{\text{ ab}}$	81.6 ± 2.92^{a}	1.3	27.2
<i>Nomia</i> sp. (n = 11)	1.3 ± 0.05 bcd	2.0 ± 0.06 ^{ab}	$74.2 \pm 3.30^{\text{ ab}}$	-0.1	13.3

Means followed by the same letter in superscript within a column are not significantly different based on Student-Newman-Keuls test at 95% significance level. Number in parentheses represents N value. OP = open pollination, BT = bagging throughout.

4.3.5 Eggplant (*Solanum melongena* L.) pollinator diversity, behaviour, effectiveness and efficiency

Six bee species were observed during the 150 observation hours visiting eggplant (*S. melongena*) flowers. The floral visitors differed significantly on the mean number of flowers visited during a foraging bout ($F_{5, 455} = 24.628$, p = 0.000) and the mean time taken per bout ($F_{5, 455} = 5.091$, p = 0.000). *Apis mellifera* and *X. calens* were more frequent but the latter visited more flowers during a foraging bout (Table 4.9).

Table 4.9: Floral visitor diversity and behaviour on the *Solanum melongena* flowers in Kakamega forest farmlands, 2009

Bee species	% Relative frequency	Mean number of visits per foraging bout \pm s.e	Mean time taken per foraging bout $(s) \pm s.e$
<i>Apis mellifera</i> (n = 213)	46.2	3.4 ± 0.44 bc	37.4 ± 3.11 ^a
<i>Xylocopa calens</i> (n = 193)	41.9	$9.7 \pm 0.46^{\ ab}$	55.5 ± 3.27 ^a
<i>X. inconstans</i> (n = 19)	4.1	12.0 ± 1.48 ^a	65.5 ± 10.43 ^a
<i>Nomia</i> sp. (n = 17)	3.7	1.4 ± 1.56 ^c	23.8 ± 11.02^{a}
Amegilla mimadvena (n = 15)	3.3	5.7 ± 1.66 bc	34.2 ± 11.74^{a}
Xy locopa flavorufa (n = 4)	0.9	4.3 ± 3.21 bc	25.3 ± 22.73 ^a

Means followed by the same letter in superscript within a column are not significantly different based on Student-Newman-Keuls test at 95% significance level. Number in parentheses represents N value.

ANOVA results indicated significant differences in fruit weight ($F_{7, 178} = 4.680$, p = 0.000) and fruit longitudinal diameter ($F_{7, 178} = 4.861$, p = 0.000). Due to the fruit characteristics of *S. melongena*, number of seeds per fruit were not counted. In this case, pollinator effectiveness and efficiency in relation to seed set was not reported but fruit weight and size were used to compare the single visits by the different pollinators. Visitations by *A. mellifera* as well as *X. calens* resulted in heavier and bigger fruits and were considered more effective compared to visitations from *X. inconstans* and *Amegilla mimadvena*. However, *Xylocopa calens* was the most efficient, spent less

time per flowers and the visit resulted into relatively heavy fruits compareable to those with unrestricted pollinator visits (Table 4.10).

Table 4.10: Mean fruit weight and length of Solanum melongena in Kakamega forest farmlands,2009

Pollination treatment/Yield parameters	Fruit weight $(g) \pm s.e$	Fruit length (cm) \pm s.e
OP (n = 47)	388.5 ± 24.59 ^a	16.8 ± 0.77 ^a
HCP $(n = 26)$	199.4 ± 33.06 ^{ab}	10.8 ± 1.03 ^a
PA(n = 16)	268.7 ± 42.15 ^{ab}	13.6 ± 1.32^{a}
BT $(n = 27)$	308.7 ± 32.44^{a}	13.7 ± 1.01^{a}
<i>Apis mellifera</i> (n = 41)	376.4 ± 26.33 ^a	16.9 ± 0.82 ^a
<i>Xylocopa calens</i> $(n = 11)$	332.9 ± 50.83 ^a	16.2 ± 1.59^{a}
Amegilla mimadvena ($n = 15$)	276.7 ± 43.53 ^{ab}	13.7 ± 1.36^{a}
<i>Xylocopa inconstans</i> (n = 3)	114.3 ± 97.33 °	10.5 ± 3.04 ^a

Means followed by the same letter in superscript within a column are not significantly different based on Student-Newman-Keuls test at 95% significance level. Number in parentheses represents N value. OP = open pollination PA = pollen augmentation, HCP = hand cross-pollination.

4.3.6 Okra (*Abelmoschus esculentus* L. Moench) pollinator diversity, behaviour effectiveness and efficiency

Flowering started 45 days from seed germination. The flowers have white-yellow petals with purplish spot at the centre and are perfect (male and female reproductive parts on the same flower). The flowers were opened from early morning 0700 hours, completely opened by 0900 hours and closed by 1400 hours of the same day (the flowers are only opened for one day). Floral visitors were observed soon after complete flower opening and continued until end of anthesis. Six bee species and a muscidae fly were recorded visiting *A. esculentus* (Table 4.11). *Lasioglossum* sp., *Apis mellifera* and were the most frequent flower visitors and important in the pollination of this crop. Muscidae flies, due to their small size relative to the okra flowers, were not considered as important floral visitors. During foraging, *Lasioglossum* sp., *Apis mellifera* and *Tetraloniella buharti* alighted on the flower petals and crawled head down to the five nectaries at the base of the petals. While imbibing

nectar from nectaries, the sticky large pollen from the anthers dusted onto the ventral and pleural body sides and on their legs. On exiting the flower, pollen was deposited on to the globolous stigma, thus encouraging self-pollination. Cross-pollination would be effected during the next successive visit. Although *Tetraloniella buharti* foraged legitimately for nectar, they were infrequent when compared to *A. mellifera* and *Lasioglossum* sp. *A. mellifera* were observed to avoid getting in touch with the large sticky okra pollen and groomed off the attached pollen from their bodies after nectar collection. They were also observed collecting nectar from the extrafloral nectar between the corolla and the calyx. An unidentified ant species were also observed collecting the extrafloral nectar.

Floral visitors took considerable time while handling these flowers during nectar and/or pollen collection. During a foraging bout *Lasioglossum* sp. visited mean of 1.0 ± 0.02 , n = 878 flowers and took longer time (167.0 s ± 5.77, n = 878), compared to *A. mellifera* that visited 1.3 ± 0.03 , n = 560 flowers while taking about 65.0 s ± 7.23, n = 560 handling okra flowers. *Tetraloniella buharti* were highly variable on the duration of their visit to 1.4 ± 0.05 , n = 130 flowers they visited. They took 20.3 s ± 15.0, n = 130 handling these flowers.

Table 4.11: Floral visitors' diversity, relative frequency of visits to *Abelmoschus esculentus* flowers in Kakamega forest farmlands, during 2009 and 2010

Bee species	Year 2009		Year 2010	
	Total visits	% Relative frequency	Total visits	% Relative
	observed		observed	frequency
Lasioglossum sp	517	57.3	361	53.5
Apis mellifera	319	35.4	241	35.7
Tetraloniella buharti	60	6.7	70	10.4
Xylocopa calens	4	0.4	-	-
Ceratina viridis	1	0.1	1	0.1
<i>Allodape</i> sp.	1	0.1	-	-
Muscidae fly	-	-	2	0.3
Totals	902	100	675	100

The yield data sets were combined for the 2009 and 2010. Flowers bagged to exclude floral visitors, left open for unrestricted visitors and the various single visits all set fruits. The results showed that okra flowers are capable of self-pollination. Nevertheless, yield results indicated

increased benefits from hand cross and insect pollination compared to spontaneous pollination. Significance differences were noted on the pod length ($F_{4, 157} = 6.160$, n = 162, p = 0.000), pod weight ($F_{4, 157} = 5.257$, n = 162, p = 0.001) and number of mature seeds per pod ($F_{4, 156} = 3.747$, n = 161, p = 0.006). However, there was minimal increase in the pod size (28%) and number of mature seeds per pod (13%) when flowers were left open to unrestricted pollinator visits. Flowers visited by *Lasioglossum* sp. gave 40% increase in the number of mature seeds compared to spontaneous self pollination. *Lasioglossum* sp. was found to be the most effective but least efficient (Table 4.12).

Table 4.12: Mean pod weight, length, number of mature seed per pod and pollination effectiveness of floral visitors on the seed set of *Abelmoschus esculentus* in Kakamega forest farmlands 2009

Bee species/Yield parameter	Mean pod weight $(g) \pm s.e$	Mean pod length $(cm) \pm s.e$	No. of mature seeds per pod \pm s.e	PEi	PEt
OP(n = 58)	18.7 ± 0.83 ^a	12.8 ± 0.38 ^a	33.9 ± 1.88 ^a	-	-
BT(n = 40)	13.5 ± 1.0^{b}	10.1 ± 0.46 ^b	29.9 ± 2.3 ^a	-	-
<i>Lasioglossum</i> sp. $(n = 38)$	18.3 ± 1.02^{a}	10.9 ± 0.47 ^a	41.9 ± 2.33^{a}	3	0.3
<i>Apis mellifera</i> (n = 18)	14.9 ± 1.49 ^{ab}	10.6 ± 0.69 ^{ab}	31.7 ± 3.38 ^a	0.5	0.6
Tetraloniella buharti (n = 8)	19.2 ± 2.23^{a}	12.6 ± 1.03 ^{ab}	33.5 ± 5.07 ^a	0.9	2.3

Means followed by the same letter in superscript within a column are not significantly different based on Student-Newman-Keuls test at 95% significance level. Number in parentheses represents N value. OP = open pollination, BT = bagging throughout.

4.4 Discussion

Spider plant (Cleome gynandra L.) pollinator diversity, behavior, effectiveness and efficiency

Nocturnal anthesis of *C. gynandra* flowers corresponded well with the nocturnal visitations by several hawkmoth species. Based on the hawkmoth's foraging behaviour, proboscis length in relation to the flower's gynophores length and the presence of pollen, the short-tongued Sphingidae *Hippotion* species seemed better pollinators. Although only 6% of *Agrius convolvuli* individuals had pollen on their proboscis, they were the most frequent and could be important pollinators as well.

The fast foraging behaviour of hawkmoths and their ability to move great distances makes them important agents for cross-pollination of many other nocturnal sphingophilous plant and crop flowers. Records of *Apis mellifera*, *Lasioglossum* sp. and other bee species are consistent with other reports that have considered bees as ubiquitous visitors to hawkmoth flowers in the late afternoon and early morning hours (Grant, 1983; Dafni et al., 1987; Cane, 2008). For example, Machado et al. (2006) indicated that bee species including Xylocopa grisescens: (Apidae) were nectar and pollen thieves while visiting flowers of Cleome spinosa. With the characteristic C. gynandra floral morphology that discourages geitonogamous pollination, these bees were unable to contact the gynophores. Contrary to this, Cane (2008) despite the nocturnal floral anthesis, reported that the most important pollinators of Cleome serrulata and C. lutea in Utah, USA were Apis mellifera and Megachile rotundata. In the current study, foraging by bees species reduced pollen flow and the amount of nectar available to the legitimate pollinators. To counter this, C. gynandra flowers compensated for the nectar and pollen compensation soon after visits by the reward thieves. It is speculated that such reductions in nectar rewards would have encouraged more visits by the hawkmoths to flowers and increased more cross-pollen movement than self-pollen thereby enhancing seed set and quality.

From the results, *C. gynandra* flowers could be considered to display a relatively moderate specialization with the hawkmoths guild since its nectar is accessible to both short and long-tongued hawkmoths. Machado *et al.* (2006) indicated that despite recording two bats (*Glossophaga soricina*: Glossophaginnae) and (*Phyllostomus discolour*: Phyllostominae) and two Sphingidae moth species (*Erinnyis ello* and *Agrius cingulata*), short-toungued sphingid moths were the most likely pollinators of *C. spinosa* in other sites. The hawkmoths species observed in this study are highly polyphagous and have been recorded visiting a diverse range of other nocturnally flowering plant species like orchids (Martins and Johnson, 2007) and crops like bottle gourd (Morimoto *et al.*, 2004) and papaya (Martins and Johnson, 2009). Papaya, an economical fruit crop in Kenya, is pollinated by *Hippotion celerio, Nephele comma* and *Agrius convolvuli* (Martins and Johnson, 2009).

The role of hawkmoth as pollinators have been underlooked in agricultural production, forests and other ecosystems, and this study highlights their direct role in agriculture and nutrition security. Pollination service by these wild hawkmoth populations may be in jeopardy especially in the tropics due to factors such as habitat fragmentation (Willot, 1999; Beck *et al.*, 2002; Schulze and Fieldler, 2003; Martins and Johnson, 2009) which have been identified as one of the factors
contributing to reductions in hawkmoth species richness and finally their pollination service. The general rareness of hawkmoths may be a challenging problem for large-scale production of *C. gynandra* seeds. In this case, hand pollination would be expensive for the seed growers and managed pollination is impractical. It is therefore necessary that research is focused on hawkmoths habitat requirements and other practises that would enhance their population, for example through habitat rehabilitation and reducing use of pesticides. This could however be challenging, considering that larval stages of various hawkmoths recorded in this study are leaf feeders and are economically important pests of agricultural crops such as *Brassicas*, tomatoes, tobacco and cut-flowers for example, roses (Kasina pers. comm.). Integrated pest management in such cases must be utilised to enhance hawkmoths adult population for improved pollination service of crops such as *C. gynandra*. Further, *C. gynandra* is no doubt a strong attractant offering both pollen and nectar to a wide range floral visitors and can serve as a good attractant to boost pollinator populations in plant community rehabilitation and restoration programmes as well as agricultural systems.

Slender leaf (Crotalaria brevidens L.) pollinator diversity, behavior, effectiveness and efficiency

Despite several bee species recorded visiting C. brevidens flowers, the pollination system of this crop could be considered more specialised. Only visits from the large-bodied solitary bees, Megachile and Xylocopa species were considered legitimate and resulted into fruit and seed set. Foraging behaviour of *Megachile* spp. and presence of the metasomal ventral scopae with unbranched hairs offered a wide area for pollen collection as well as contact with the stigmatic surface. *Xylocopa* spp. contact with the stigma was only limited to a small area on the first metasomal segment but their heavy weight as well as their foraging behaviour increased the efficiency with which they tripped flowers. *Megachile* spp. and *Xylocopa* spp. visited many flowers per foraging trip and spent considerably longer time during these trips but the latter spent slightly more time per flower and had higher pollinator effectiveness. The higher pollinator effectiveness for Xylocopa *calens* compared to *Megachile rufipes* may be attributed to their heavy weight because of their large size, meaning that during flower manipulations, more pressure was exerted on the keel hence more pollen was dispensed and carried over on their lower abdominal segment to the next flower. It is possible that the duration of the visit to a flower influenced the efficiency of pollen transfer and the resultant seed set. Contrary to this, further results indicated Megachile rulipes to be the most efficient (setting more seeds per unit second of visit). Generally, as the duration of a visit to a flower increased, so was the number of pressings on the keel and exposure of the reproductive structures hence more rubbing and abrasion of the stigmatic membrane and cross-pollen deposition. Such a relationship may not necessarily hold, if longer duration of visit means additional pressings that may inflict cumulative damage on the flower. Cumulative effects on seed set in relation to the number of keel pressings made by a pollinator during a visit was not evaluated in the current study but will be of interest in future studies.

While considering the structural features of the legitimate pollinator, the floral morphology of the study crop and pollinator abundance, Megachile spp. may be considered as specialised pollinator of C. brevidens at the study site. On the other hand, Thyreus spp. recorded on C. brevidens flowers though considered inconsequential to pollination of the crop, appeared unique. Mostly, they are regarded as nest parasites of other anthophorid bees and lack pollen-collecting structures (Michener, 2000). Still, their record and interactions with other floral visitors could have had an effect on the behaviour and effectiveness pollination by other legitimate visitors. From the data on the pollinator effectiveness, leafcutter bee, Megachile rufipes is the most effective pollinator. They were also the most frequent and should therefore be considered important for seed production of this crop. It would therefore be important to explore the possibility of domesticating Megachile rufipes in artificial nests such as drilled wooden or polystyrene blocks. Leafcutter bee, Megachile rotundata, has been successfully used for managed pollination of alfalfa in the USA and Canada. Their management is based upon providing nesting sites such as solid nesting boards, or laminated polystyrene blocks and then introducing them into flowering alfalfa seed fields (Delaplane and Mayer, 2000). Habitat fragmentation, pesticide use and agricultural intensification are some of the threats to the abundance and diversity of these bees. Evaluation of integrated pest management strategies that would increase seed production and reduce pesticide use are recommended. Alternatively, seed production sites could be located in areas with either Megachile or Xylocopa spp. These areas include among others, the farmlands neighbouring Kakamega forest where over 30 Megachile spp. have been recorded on C. brevidens (Gikungu, 2006).

Karela (Momordica charantia L.) pollinator diversity, behavior, effectiveness and efficiency

Results from this study indicated differences in floral display between the staminate and pistillate flowers of *M. charantia*. The pistillate flowers are rewardless and are likely to be pollinated

by deceit. The long period of overlap of flowering of both flower sexes plus the numerous staminate flowers enhances the pollen flow and the overall pollination of the crop. *Lasioglossum* sp., *Apis mellifera, Plebeina hildebrandti* were considered potential pollinators of *M. charantia* in Kakamega, Western Kenya. These flower visitors clearly discriminated against the rewardless pistillate flowers. Proportion of pistillate flowers that received visitations from flower visitors were low representing 1% of all the 3693 visitations by floral visitors. It is possible that once the flower visitors experienced the rewardless pistillate flowers, second time foragers avoided these flowers in their subsequent visits all together and that pollination of this crop is purely by deceit.

Reproductive success of *M. charantia* was pollen limited. This can be attributed to the high discrimination against the pistillate flowers that would have otherwise developed into fruits that is, in case, they received visits from pollen loaded flower visitors. Flowers receiving hand crosspollination resulted in 20% higher fruit set in 2009 and 30% in 2010 while supplemental hand crosspollination increased fruit set by 14% in 2009 and 40% in 2010 compared to open pollinated (control) flowers. Pollen augmentation did not result into increased fruit set when compared to hand cross-pollination. Given the high discrimination against the pistillate flowers, it is possible that the treated pistillate flowers in this treatment did not receive any additional visits from flower visitors or the amount of pollen that was deposited on the stigma by hand was definitely enough for highest possible fruit set. Also, since the amount of pollen dusted onto the stigma was not counted, it is possible that too much pollen was deposited leading to stigma pollen clogging and reduced fruit and seed set. On the other hand, low fruit set in the open pollination treatment for 2010 compared to 2009 was attributed to the differences in pollen flow in both years. For instance, there was a high ratio of pistillate to staminate flowers (1:430 in 2009 compared to 1:227 in 2010). This meant that in 2009, there was more pollen available to flower visitors during foraging and that enough was deposited on the pistillate flowers hence better fruit set. It is believed that higher number of the staminate flowers enhances the chance of effective pollination, resulting in high fruit and seed set. However, there is need to evaluate the optimal ratio of pistillate to staminate flowers for maximum fruit production.

Fruit set results indicates for monoecious crops with nectarless pistillate flowers, for example, *M. charantia* are not suited for large scale production of fruits. Even under small scale production, supplemental hand cross-pollination is necessary for the production of marketable fruits. The poor fruit set in this study agrees with results by Mishra and Sahoo (1983) who reported fruit set of 22% in *M. charantia* in India under natural pollination conditions. On contrary, results by Deyto

and Cervancia (2009), showed high fruit set in natural pollination (78%) and there was no significant difference with hand pollinated flowers (80%). Although not mentioned in the study, the high fruit set in natural population in the Philippines by Deyto and Cervancia (2009) could have been as a result of high abundance of stingless bees Trigona and Halictus sp. in the test plot. Under natural pollination conditions, it's assumed that every flower is visited more than once and probably by many different bee species and will have a better fruit and seed set but this was not the case for M. charantia in the study area. Poor yields recorded were consistent with observations on low visitation rates to the pistillate flowers. In agricultural production, where higher fruit set, bigger fruits with more uniform shapes are desired, two options are possible to increase the fruit set of *M. charantia*, (1) the introduction of managed (stingless) pollinator populations e.g. A. mellifera, P. hildebrandti, (2) farmers must resort to hand pollination to produce any marketable fruits. Indeed, other authors have recommended hand pollination for commercial crop and seed production (Behera et al., 2010; Joseph, 2005; Devadas and Ramadas, 1993; Mishra and Sahoo, 1983). Devadas and Ramadas (1993) calculated that 29 man-hours were needed to produce 1 kg of commercial seeds. Hand pollination requires labour resource input and is an added cost to the already resource poor farmers. Also, hand pollination may not necessarily result into better fruit set because of pollen clogging due to too much pollen deposited on the stigma. A. mellifera, Lasioglossum sp., and Plebeina hildebrandti, showed a high efficiency of pollen transfer as depicted by the high fruit set in the single visit compared to other pollination treatments. It was also evident that native bee communities were unable to offer sustainable pollination service. Therefore, pollination augmentation or managed crop pollination should form core inputs considered in the production of this crop. Increasing the density of A. *mellifera* populations by introduction of hives into the flowering crop of *M. charantia* might result into competition for nectar and pollen forcing the bees to visit even the female flowers. More research is however needed to evaluate if the introduction of honey/stingless bee hives into the crop results into less pistillate flower discrimination and higher fruit yields. Lasioglossum sp. recorded visiting *M. charantia* in this study, nests in soil and other pithy stems of herbs and shrubs (Gikungu, 2006). Observation of *Plebeina hildebrandti* visiting flowers of *M. charantia* is in particular of much interest and highlights the potential of stingless bees for crop pollination of commercially important crops (Heard, 1999; Slaa et al., 2006; Cortopassi-Laurino et al., 2006). Farming practices such as maintaining of diverse hedges around farmland landscapes increases the abundance of such pithy stems thereby enhancing the nesting sites for these bees. P. hildebrandti nests in occupied termite mounds at the core of the mound (Michener, 2000). However, termite mounds as well as the termites themselves are in most cases are cleared off from the farmlands using pesticides. Such practices may

accelerate the decline in abundance of these pollinators thereby reducing crop fruit and seed set. Further research on the potential of stingless bees and solitary bees as manageable crop pollinators will be of interest.

Broad-leafed African nightshade (Solanum scabrum Mill) pollinator diversity, behavior, effectiveness and efficiency

Solanum scabrum was exclusively visited by wild bees; especially the soil nesting Lasioglossum sp., Amegilla langi, Nomia sp. Apis mellifera were also recorded visiting the flowers. Although not capable of buzz pollination a requirement for such solanaceous flowers with poricidal anthers, they were observed vigorously striking the anthers with their forelegs to release pollen which they packed onto their corbiculae. Collection of pollen by *A. mellifera* from flowers with poricidal anthers buzz-pollinated flowers had been described by Cane *et al.* (1993) and Thorp (2000). They referred to this behaviour as "drumming" or "milking" of poricidal anthers to release pollen. There was only 6% increase in seeds per berry under open pollination over self-pollinated berries thus indicating a plant capable of seed set without pollinators. Amongst the bee pollinators, *Amegilla langi* and *X. calens* were the most effective and efficient pollinators but they visits resulted into number of seeds that were compareable to those flowers receiving unrestricted pollinator visits. Pollination management for this crop may not be necessary.

Eggplant (Solanum melongena L.) pollinator diversity, behaviour, effectiveness, and efficiency

Solanum melongena is a self fertile and self-pollinating plant; cross-pollination through insects is possible and desirable. The process of pollen release from the poricidal anthers onto the stigma requires shaking either by wind or insect visitors (Free, 1993). While considering fruit size and weight, visitations by pollinators resulted into 26% heavier fruit compared to when pollinators were excluded. Perharps, due to considerably large flowers, wind agitation of the flowers is not sufficient to release pollen from the poricidal anthers. In the current study, *Apis mellifera* and *Xylocopa calens* were the most frequent with over 40% relative frequency of the total observed floral visitors. Fruit weight of the flowers they visited were compareable to those that received unrestricted

pollinator visit. They were therefore considered effective pollinators of eggplant in the study area. *A. mellifera* are not able to buzz-pollinate (Buchmann, 1983), but their visitation resulted into fruit weights that were 13% heavier compared to *X. calens* visits. Their characteristic "milking" and "drumming" while foraging on flowers with poricidal anthers have been described by Cane *et al.* (1993) and Thorp (2000). In a study in Nguruman area in Kenya, Gemmill-Herren and Ochien'g, (2008) found that, *Xylocopa caffra* and *Macronomia rufipes* were the most effective. Amoako and Yeboah-Gyan (1991) in a study in Ghana indicated that visitations by honeybees (*A. mellifera adansonii*) resulted into increased eggplant yields. Other results include those by Levin (1989) who also found out that *A. mellifera* visitations improved the eggplant fruit weight. Gemill-Herren and Ochien'g (2008) noted that the number of seeds was positively correlated to the fruit weight. Due to the eggplant fruit fleshy characteristics, it was not possible to count the number of seeds per fruit but future evaluation of pollinator effectiveness such fleshy fruits should consider (1) letting the fruit samples to rot, (2) washing and straining the seeds using a siever, and (4) finally counting them.

For managed pollination for this crop, farmers are encouraged to leave patches of uncultivated lands with flowering plants near their farmlands that would provide alternative nectar and pollen resources to the pollinators. Alternative ruderal flowering weeds, forest habitats and riverine woodlands, enhance bee species diversity and abundance in the nearby eggplant farms (Gemmill-Herren and Ochien'g, 2008), thus improving pollination, fruit set and quality.

Okra (Abelmoschus esculentus Moench) pollinator diversity, effectiveness, and efficiency

Although self-pollination readily occurs in okra, the flowers are attractive to various floral visitors. *Lasioglossum* sp. was the most frequent and effective in the pollination of this crop. They foraged legitimately and took longer time collecting nectar and pollen when compared to other floral visitors. It is believed that, the longer they stayed on the flower, the more pollen was dusted onto their bodies as they successively went from one nectary to another while collecting nectar. *A. mellifera* comparably took shorter time and were observed to clean off attached okra pollen from their bodies. This behavior has been recorded on *Gossypium* flowers and was attributed to the large sizes of these pollen grains (Buchman and Shipman, 1990). In a study on the pollen sources for *A. cerana* and *A. mellifera* in India, Suryanarayana *et al.* (1992) noted that *Abelmoschus* sp. pollen was not recorded despite these bees readily visiting these flowers for nectar. Vaissiére and Vinson (1994)

found that the presence of spines in *Gossypium* sp. and *Abelmoschus* sp. pollen interfered with the normal pollen collection and packing resulting into low pollen harvesting efficiency. They also noted that A. mellifera were unable to pack Abelmoschus esculentus pollen due to their very large Equivalent Spherical Diameter (ESD) of 155.7 µm and the long spines (23.8 µm). Although the pollen size was not measured in the current study, the pollens were observed to be large and sticky. It is presumed that A. mellifera avoided them most likely due to the above morphological characteristics. Okra is self-pollinating and would set fruit even in the absence of pollinators. But, pollinators improved fruit yield. Unrestricted pollinator visits increased the pod size by 28% and number of mature seeds per pod by 13% when compared to spontaneous self-pollination while those flowers visited by Lasioglossum sp. gave 40% increase in the number of mature seeds compared to spontaneous self-pollination. These results agree with those recorded by Njoya (2005) who recorded 22% increase in seed sets by insect pollination compared to spontaneous self-pollination. He further considered *Halictus* sp. and *Megachile* sp. important pollinator of okra in Cameroon. In this study, these bees were not recorded visiting okra. Tetraloniella buharti though infrequent, was the most efficient due to the lesser time spent per flower during foraging. Nevertheless, Spear's pollinator effectiveness revealed that pollination effectiveness for A. mellifera was low (0.5 seeds per single visit) compared to *Lasioglossum* sp. (3 seeds per single visit). The behavior of okra pollen avoidance by Apis mellifera is likely to reduce the amount of pollen flow and negatively affect their effectiveness as pollinators, and in general the pollination of this crop. Lasioglossum sp. was therefore the most frequent and effective pollinator for okra at this study site.

4.5 Conclusions

In summary, the results presented here highlight the importance of wild pollinators in crop production. It is further expected that if more such studies are carried out for other crops, the most effective pollinators are likely to be other pollinators other than honeybees. Also, the study provided the first quantification of the pollination effectiveness of floral visitors for various economically important vegetables, spider plant (*Cleome gynandra*), slender leaf (*Crotalaria brevidens*), broadleafed African nightshade (*Solanum scabrum*), karela (*Momordica charantia*) and okra (*Abelmochus esculentus*). However, these floral visitors may differ in other localities from those observesd in this study. Factors likely to affect the pollinator effectiveness include the visitation rates of the pollinators

which may vary with the presence of competing flowers for the same pollinator, time of day and other environmental variables such as temperature. In addition, relative abundance of the floral visitors may vary among years and cropping systems. With the global decline of the managed honeybee populations due to the widespread of the Varroa mite and the Colony Collapse Disorder (CCD), wild bees may provide the much needed insurance from reduced pollination. *Varroa destructor* has recently been reported in many honeybee hives across East Africa (Faizer *et al.*, 2009). Research and conservation of wild bee population is emphasized for sustainable crop pollination.

5. POTENTIAL OF HONEYBEE (APIS MELLIFERA L.) FOR POLLINATION GREENHOUSE TOMATOES (LYCOPERSICON ESCULENTUM MILL) IN KENYA

5.1 Introduction

Tomatoes (*Lycopersicon esculentum* Mill.), have poricidal anthers from which pollen is released when the androecium or anthercone is shaken (McGregor, 1976). In the field, pollination is achieved by wind agitation that usually triggers pollen release, promoting self-fertilization or by occasional insects that encourage cross-pollination (Free, 1993). In order to obtain the tomato pollen, insect pollinators must grip the ends of the anthers and vibrate their wing muscles, leading to dislodging of the pollen grains, an activity termed as 'buzz' pollination (Buchmann, 1983). However, in the greenhouse, wind and insect pollinators are absent and successful pollination and fruit set is difficult. Banda and Paxton (1991) found that fruit set of tomatoes grown in greenhouses is frequently poor and is highly dependent on some mechanical aids. Various techniques have since been adopted to increase fruit set and yield in such enclosed environment. These include mechanical shaking of the plant or individual flower clusters either by hand or electric vibration wand, and introduction of insect pollinators into the greenhouses (Banda and Paxton, 1991; Dogterom *et al.*, 1998; Morandin *et al.*, 2001a). Other than fruit set, insect-facilitated pollination have also been shown to increase fruit size (Higo *et al.* 2004; Sabara *et al.*, 2004).

Several bee species have been evaluated and are currently used for pollination of greenhouse tomatoes. For example, honeybees, *Apis mellifera* (Banda and Paxton, 1991; Cribb *et al.*, 1993; Sabara and Winston, 2003; Higo *et al.*, 2004; Sabara *et al.*, 2004), bumblebees, *Bombus* spp. Latreille (Banda and Paxton, 1991; Dogterom *et al.*, 1998; Morandin *et al.*, 2001a, b) have been reported to successfully pollinate tomatoes under such conditions. In Australia, solitary bees that perform buzz pollination, such as *Amegilla chlorocyanea* and *A. (Zonamegilla) holmesi* (Anthophoridae), and *Lestis aeratus* and *L. bombylans* (Apidae: Xylocopinae) have also been found to be very efficient in pollinating greenhouse tomatoes (Hogendoorn *et al.*, 2000; 2006; Bell *et al.*, 2006). Recently stingless bees, *Nannotrigona perilampoides* (Hymenoptera: Meliponini) in Mexico (Cauchi *et al.*, 20, 2005).

2004; Palma *et al.*, 2008) and *Melipona quadrifasciata* (Hymenoptera: Apidae) in Brazil (Del Sarto *et al.*, 2005; Bispo dos Santos *et al.*, 2009) have been investigated for pollination of tomatoes and other crops including green peppers. Among the bee pollinators, reared colonies of *Bombus impatience*, *B. occidentalis*, *and B. terrestris* are currently exported worldwide and used especially in Europe and USA for pollination of tomatoes and many other crops (Delaplane and Mayer, 2000).

No research study has been undertaken to evaluate the potential of honeybees or any other bee species for greenhouse tomato pollination in Kenya. This is despite the rapid spread of tomato production under greenhouse. One of the major problems in the greenhouse tomato production is insufficient pollination due to the enclosed conditions and limited wind, resulting in low production and poor quality fruits. In addition, there is also lack of information of the type of crops to be grown under enclosed conditions. Crops that have been planted in such conditions without successful fruit set include watermelon and courgette (pers. observ.). There are also no native bumblebee species and farmers in the near future may lobby for the importation and use of bumblebees for pollination of greenhouse tomatoes as well as other crops. Importation of commercial tomato pollinators such as the bumblebees could pose a considerable danger to indigenous bee fauna in the country. Importations of exotic bees such as bumblebees into new areas have been shown to present environmental problems such as proliferation of weeds and other alien plant species through enhanced seed set (Hanley and Goulson, 2003). There is need to find alternative native bee species that can be utilized to improve fruit set and quality of tomatoes under enclosed conditions. Other research on the use of honeybees for pollination of greenhouse tomatoes have been undertaken in different environmental conditions while using other tomatoes varieties. With the declining diversity and abundance of both wild and managed pollinators, stingless bees have increasingly been recognised as potential crop pollinators (Heard, 1999; Slaa et al., 2006; Cortopassi-Laurino et al., 2006). They are social, lack functional sting, collect both nectar and pollen for the brood and live in permanent colonies (Michener, 2000). The aim of this study was to evaluate honeybees (Apis mellifera L.: Apidae), for pollination of two common greenhouse tomatoes varieties (Chonto F1 and Plumpty F1) in Kenya. Stingless bees species, Meliponula bocandei and Meliponula ferruginea (Hymenoptera: Apidae) were also evaluated for pollination of tomatoes under greenhouse.

5.2 Materials and methods

5.2.1 Study sites

The experiment was conducted at two separate sites, (1) experimental site located within the Kenya Agricultural Research Institute, National Agricultural Laboratories (KARI-NARL) in Nairobi and (2) commercial site in Ongata Rongai, Nairobi. At KARI-NARL, three glasshouses of 4 m x 3 m were planted (11th March 2010) with tomato variety Chonto F1 (blocky in shape) seedlings raised in 16 inch poly-sleeves filled with sterilized soil at a spacing of 60 cm x 30 cm (66 plants per glass house). At the commercial site, a 30 m x 8 m greenhouse covered with polythene was planted (10^{th}) June 2010) with tomato variety Plumpty F1 (oval-shaped) seedlings at a spacing of 30 cm x 30 cm (2,100 plants) on raised beds (90 cm wide with 30 cm between beds). Since honeybees foraging distance was confined within 10 m from the location of the hive, the last 20 m of the greenhouse full length and two beds along the greenhouse walls (one bed on each side of the greenhouse) were excluded from the experiment. Crop management at both sites was done as specified in KHDP (2008). Watering at KARI-NARL site was done by hand and 500 ml was given to each plant after every 2 days while at the commercial site, watering consisted of drip irrigation. No automated temperature or humidity controls were used at both sites and so conditions inside depended on the outside weather conditions. In both cases ventilation was through the top vents that remained opened throughout the study period, and as a result, bees were able to exit and return to the greenhouse. Relative humidity and temperature inside were measured using Data loggers (Tiny Tag®).

5.2.2 Bee colonies

Stingless bee colonies of *Meliponula ferruginea* and *M. bocandei* maintained in modern hives were collected from Kakamega (BIOTA-E10 subproject and Mr. Stanley Imbusi, a renowned farmer practising meliponiculture), while two *Apis mellifera* housed in Langstroth hives were sourced from the KARI-NARL apiary. The stingless bee colonies were relocated from Kakamega and allowed 2 weeks of acclimatization period at the KARI site. The bee colonies, *Meliponula bocandei*, *M. ferruginea* and *Apis mellifera* were introduced on the 3rd May 2010 into the three glasshouses; A, B and C respectively. At the commercial site in Rongai, only *Apis mellifera* was introduced on the 7th October 2010. The hives were placed at the furthest end of the greenhouse on

platforms raised 1 metre from the ground and left to acclimatize to the greenhouse environment for 3 days. All introductions and removals of the hives were done late in the evening at 1900 hours after all the foragers had returned to the hive. Since tomato flowers do not provide nectar to insect foragers, nectar supplement was offered to bees in form of honey-water at the ratio of 2 *Apis* honey: 1 tap water placed in blue coloured plates placed in each glasshouse (Dogterom *et al.*, 1998; Dogterom and Winston, 1999). Additional clean tap water was offered to bees in each greenhouse. The supplemental honey-water and tap water were replaced every 3 days. Pieces of small dry wooden sticks were dropped into the honey-water and water to act as landing platforms to facilitate collection without the bees drowning.

5.2.3 Bee foraging activity and colony traffic

Bee forage activity at KARI-NARL was observed through the see-through glass walls while at the commercial site Rongai, observations were made by greenhouse walk-throughs. The number of workers of *M. bocandei*, *M. ferruginea* and *A. mellifera* foraging or flying in the respective glasshouses were counted and recorded every hour from 0800 to 1600 hours. Additional information on the number of flowers visited in a foraging bout by an individual bee and the duration (seconds) of the foraging bout were recorded. To evaluate the colony traffic, the numbers of foragers leaving the hive and returning to the hive within 5 minutes every hour from 0800 to 1600 hours were also noted. To determine how readily the bee species accepted supplementary honey-water offered, the number of bees collecting the honey-water and water were counted every hour from 0800 to 1600 hours. Effect of greenhouse temperature (°C) and relative humidity (%) on the colony traffic as well as the number of foragers inside the greenhouse was determined.

5.2.4 Effect of pollination treatments on tomato fruit set and yield parameters

Pollination treatments were limited to honeybees, as stingless bees did not visit tomato flowers. Pollination treatments included (1) bagging (BT), (2) open without honeybees (OP), (3) manual truss shaking (MTS), (4) (HB) single visit by honeybees (HBsv), (5) and multiple visits (HBmv). In bagging, flowers were bagged throughout until fruit set. Single visit treatment (HBsv) involved exposing, previously bagged flowers to bee foragers until they received a single visit, and then bagged, while in multiple visits, flowers that received more than a single honeybee visit (>1 visit) were noted. For MTS, flower trusses (8 flowers) were randomly selected and an evening before opening they were bagged using fine organza bags. The following day after flower opening, MTS treatment was performed by hand tapping of individual flower trusses using fine forceps until a cloud of pollen was observed around the flowers and then the flowers re-bagged until fruit set. Each truss was shaken for 1 minute thrice a week between 1000 hours to 12 hours. In Kenya, some greenhouse tomato farmers shake the whole or tap the trellising wires from where the tomato plants are trained (pers. observation). The tappings of the trusses were considered effective when a cloud of pollen dust was visible from the anthers. MTS were repeated for three consecutive days. This method, however, was believed to aid self-pollination than cross-pollination. The bagging material were removed 15 days after flower manipulations, and fruit set recorded across all the treatments. Fruits were harvested when completely orange-red and weighed immediately to the accuracy of 0.01g using Mettler Toledo 802E precision balance. The fruit length (vertical length) and maximum diameter (transverse circumference) of the tomatoes were measured using a digital vernier calliper to the accuracy of 0.01 mm. The fruits were then individually pressed and passed through a sieve and the number of seeds counted per fruit for each treatment. The variables measured included, percent fruit set, fruit weight, length and maximum diameter, number of seeds and weight of dry seeds per fruit.

5.2.5 Data analysis

Percent fruit set was calculated as then number of fruit set per truss divided by the number of flowers treated per truss (8 flowers) x 100. Data on colony traffic (number of bees leaving and returning to the hive), foraging behaviour (number of foragers in the greenhouse and the number of flowers visited per bout and the duration of a foraging bout), and yield parameters (fruit weight, length, maximum diameter, number of seeds and weight of dry seeds per fruit) were subjected to Univariate Analysis of Variance (ANOVA) using General Linear Model procedure (IBM SPSS® version 19) at 95% significance level. *Post hoc* tests using Student-New-Keuls were performed to separate the means whenever significance differences were indicated by GLM procedure. Pearson correlation coefficients of temperature (°C) and relative humidity (%) with colony traffic and number of honeybee foragers in the greenhouse, number of seeds per fruit per fruit with weight of fruit, height and maximum fruit diameter were performed. The results were considered significant when p ≤ 0.05 , highly significant if p ≤ 0.001 and marginally significant if $0.10 \ge p \le 0.05$).

5.3 Results

5.3.1 Bee foraging activity and colony traffic

Meliponula bocandei and Meliponula ferruginea foraging activity and colony traffic

Soon after introduction, both *M. bocandei* and *M. ferruginea* closed the hive exit using batumen. They were also observed to close the hive exits early mornings and late evenings. Foragers of *M. bocandei* started to exit the hive the next morning after introduction into the glasshouse. However, these bees were not seen foraging on the tomato flowers or standing on the plants leaves but were observed crawling against the glass wall to the Eastern direction of the sunrise. They neither collected the supplemental honey water nor water offered. Results on the colony traffic indicated that the total number of individual bees that exited the hive per day were higher than the number that returned to the hive colony (Figure 5.1a). Most of the bees that could not locate the hive died after 2-3 days. This reduced the colony foraging strength. The colony became inactive (very few individuals exiting) and on opening of the hive, many adult individuals had died and there was neither a live adult nor larvae. The colony finally collapsed after 3 weeks of introduction (n = 23 days). On the other hand, M. ferruginea, were able to survive the enclosed environment during the observation period (n = 27 days) but they were never observed visiting flowers or collecting the supplemental honey water or water offered to them. They were seen peeping from the hive exit, with very few bees leaving the hive. Though the number of foragers exiting per day was higher than the total numbers returning to the colony per day, the colony remained active for the entire observation period (Figure 5.1b). They located the glasshouse vent and flew out to collect other pollen and nectar from alternative flowering plants occurring outside the glasshouse. Adult bees were observed when the hive was opened. The colony resumed activity once they were removed from the glasshouse environment. The evaluation of *M. bocandei* and *M. ferruginea* as possible pollinators of greenhouse pollination of tomatoes was terminated, as they were considered ineffective.

Foraging behaviour and colony traffic of honeybees (Apis mellifera)

Apis mellifera were observed visiting tomato flowers. The foragers were observed packing pollen onto their corbiculae. To dislodge the pollen, the bees were observed to use their forelegs to vigorously strike the poricidal anthers to release pollen. They took exceptionally long time on these

flowers more than 190 seconds on Chonto F1 flowers at KARI-NARL experimental site, and about 150 seconds at commercial site, Rongai. During the foraging bout approximately 3.0 ± 0.28 , n = 32 flowers and 2.0 ± 0.11 , n = 39 were visited at the different sites respectively.



Figure 5.1: (a) Daily colony traffic of *Meliponula bocandei*; (b) daily colony traffic of *Meliponula ferruginea*; (c) mean daily temperature and relative humidity in glasshouse with *M. bocandei*; and (d) mean daily temperature ($^{\circ}$ C) and relative humidity in glasshouse with *M. ferruginea*

KARI-NARL site

Apis mellifera were observed to be erratic for first days after introduction in the small glasshouse. Many foragers were seen on the glasshouse walls during the early part of the morning. They were able to return to the hive by late evening. They readily accepted the honey-water solution but did not collect the water placed in the blue coloured plastic plates. A total of 46 individual bees were observed taking honey water and one individual taking water by the 4th day of introduction with no further observation of honey-water or water collection. On the 13th day, a new colony was brought

in and the erratic behaviour was repeated (1st day of the second colony). By the 2nd day, 90 individual bees were counted collection honey-water. Honey-water collection occurred between 1000 to 1500 hours. Glasshouse temperature and humidity were significantly different ($F_{8, 147} = 46.584$, p = 0.000; $F_{8, 147} = 4.115$, p = 0.000) respectively with low temperatures being registered early in the morning compared to late afternoons. Mean number of honeybee foragers in the glasshouse was not significantly different across the day (p > 0.05) with a mean of 36.2 ± 3.98 , n = 19, individual bees observed in the glasshouse (Figure 5.2).



Figure 5.2: Mean (\pm s.e) of relative humidity (%), temperature (°C) and number honeybee foraging across the day, experimental site, KARI-NARL, 2010. Bars are standard error of the mean.

There was marginal significant difference on mean numbers of foragers leaving the hive across the day (p = 0.09) with a mean of 1.7 ± 0.18 , n = 156 individual bees leaving the hive every 5 minutes. Honeybees were able to return to the hive but the number of forages returning to the hive were significantly different (F _{8, 147} = 2.955, p = 0.004) across the day (mean 1.9 ± 0.16 , n = 156). Fewer foragers returned to the hive at 0900 hours compared to 1100 hours (Figure 5.3).



Figure 5.3: Mean (\pm s.e) number of honeybees leaving and returning to hive during 5 min every hour at experimental site, KARI-NARL. Bars are standard error of the mean.

Commercial site (Rongai)

Temperature and humidity were significantly different across the day ($F_{8, 189} = 87.44$, p = 0.000) and ($F_{8, 189} = 23.45$, p = 0.000) respectively. Higher relative humidity was recorded in the early morning but this reduced across the day (Figure 5.4). The number of foragers leaving and returning to the hive during 5 minutes every hour was significantly different across the day ($F_{8, 189} = 3.442$, p = 0.001) and ($F_{8, 189} = 2.664$, p = 0.009), respectively. The hive was more active in the afternoon than morning periods. For example, fewer bee foragers left and returned to the hive in the morning compared to afternoon (Figure 5.5).



Figure 5.4: Mean (\pm s.e) of relative humidity (%), temperature (°C) and number of honeybee foraging across the day, Commercial site, Rongai, 2010. Bars are standard error of the mean.



Figure 5.5: Mean number $(\pm s.e)$ of honeybees leaving and returning to hive during 5 min every hour at Commercial site, Rongai. Bars are standard error of the mean.

Influence of temperature and humidity on honeybee (Apis mellifera) colony traffic

Environmental factors, temperature, and humidity did not influence the colony traffic at the experimental site, KARI-NARL. At the commercial site in Rongai on the other hand, colony traffic was slightly influenced by the greenhouse temperature but not relative humidity (Table 5.1). The number of individual bees leaving the hive and returning to the hive every 5 minutes per hour increased with increasing temperatures across the day (Table 5.1). The number of bee foragers collecting water and honey-water, occurred between 1000 hours and 1600 hours, and were not significantly different (p > 0.05) across the day. Neither temperature nor relative humidity influenced both water and honey-water collection (p > 0.05) by the foragers.

Table 5.1: Pearson correlation coefficients of temperature ((°C) and relative humidity (%) with colony
traffic and number of honeybee foragers in the greenhouse,	, 2010

Variables	Experimental site	erimental site <i>P-value</i>		<i>P</i> -value
	(KARI,NARL) (r)	(Rongai) (
Foragers in the house				
Temperature	0.084	0.295	0.025	0.727
Relative humidity	0.039	0.631	0.062	0.389
Bees leaving the hive				
Temperature	0.073	0.368	0.150	0.035*
Relative humidity	-0.025	0.756	-0.043	0.550
Bees returning to the hive				
Temperature	0.149	0.063	0.158	0.026*
Relative humidity	-0.024	0.766	-0.024	0.740
Ν		156		198

*Significant at 95% significance level.

5.3.2 Effect of pollination treatments on tomato fruit set and yield parameters

Experimental site, KARI–NARL (variety Chonto F1)

At the Experimental site, KARI-NARL, there was no significant difference (p > 0.05) in the tomato fruit weight, fruit height, and fruit maximum diameter between the pollination treatments.

Number of seeds per fruit were significantly different ($F_{4, 233} = 2.351$, p = 0.055) with a mean of 58.55 ± 3.02 seeds per fruit. The following results will therefore focus on the fruit set and yield parameters at the commercial site, Rongai.

Commercial Rongai site (tomato variety Plumpty F1)

Fruit set (%) per truss (8 flowers) was highly significantly different ($F_{3, 132} = 26.8$, p = 0.000). Percent fruit set was low (43%) in truss flowers that were bagged (BT) and open without honeybees (OP) (61%) when compared to those that were manually shaken (MTS) (76%) and the flowers that were left open with honeybees (HB) (78%), (Figure 5.6).



Figure 5.6: Mean fruit set (%) (\pm s.e) per truss (8 flowers) resulting from Pollination treatments, BT = bagging, n = 33, OP = open without honeybees, n = 36, MTS = Manual truss shaking, n = 36, HB = open with honeybees, n = 36. Bars with same letter show no significant difference from each other at 95% significance level. The error bars show the standard error.

Pollination treatments had significant effect on the fruit weight ($F_{4, 224} = 15.775$, p = 0.000), seed number per fruit ($F_{4, 224} = 51.409$, p = 0.000), fruit length ($F_{4, 224} = 20.541$, p = 0.000), fruit maximum diameter ($F_{4, 224} = 21.155$, p = 0.000) and weight of dried seeds per fruit ($F_{4, 224} = 6.799$, p = 0.000). Flowers that received facilitated pollination from honeybees (HB) and manual truss shaking

(MTS) resulted in higher fruit weight, fruit length, maximum diameter, number of seeds and weight of dried seeds per fruit when compared to flowers that received no facilitated pollination (bagged and open without honeybees) (Table 5.2). Flowers that received single visit by honeybees (HB) were 27% heavier and 55% higher seed number per fruit compared to flowers that received no facilitated pollination. Further results indicated that there was no increase in tomato fruit weight, number of seeds, length, and maximum diameter with additional honeybee visits to flowers (Table 5.2). Pearson correlation (2-tailed test) showed that there was a high positive correlation between the number of seeds per fruit with the weight of fruit (r = 0.690, p = 0.000, n = 229), fruit length (r = 0.574, p = 0.000, n = 229) and maximum diameter (r = 0.700, p = 0.000, n = 229).

Table 5.2: Comparison of mean (\pm s.e) of weight, seed number, length, maximum diameter and dry seed weight per fruit from no facilitated pollination (BT and OP), manual truss shaking, single visit and multiple visits from honeybees pollination, commercial site, Rongai, 2010

Yield parameters	BT	OP	MTS	HBsv	HBmv
	$(n = 33)^{a}$	(n = 36)	(n = 36)	(n = 67)	(n = 57)
Weight of fruit (g)	$45.2 \pm 3.45^{\circ}$	53.9 ± 3.30^{b}	67.9 ± 3.30^{a}	73.9 ± 2.42^{a}	70.4 ± 2.62^{a}
Fruit length (mm)	$48.2 \pm 1.30^{\circ}$	52.4 ± 1.24^{b}	53.9 ± 1.24^{b}	59.4 ± 0.91^{a}	60.5 ± 1.0^{a}
Max. diameter (mm)	$36.9 \pm 1.03^{\circ}$	$40.2 \pm 0.98^{\mathrm{b}}$	$46.2 \pm 0.98^{\mathbf{a}}$	46.8 ± 0.72^{a}	$45.1\pm0.78^{\mathbf{a}}$
No. of seed / fruit	31.3 ± 4.57^{b}	39.6 ± 4.38^{b}	94.9 ± 4.38^{a}	89.6 ± 3.21^{a}	85.1 ± 3.48^{a}
Dry seed weight/ fruit (g)	0.1 ± 0.04^{b}	0.2 ± 0.04^{b}	$0.3 \pm 0.04^{\mathbf{a}}$	0.4 ± 0.03^{a}	$0.3 \pm 0.03^{\mathbf{a}}$

Means followed by the same letter in superscript within the same row are not significantly different based on Student-Newman-Keuls test at 95% significance level. Pollination treatments, BT = bagging, OP = open without honeybees, MTS = Manual truss shaking, HBsv = single visit by honeybees and HBmv = multiple honeybee visits (>1). Number in brackets indicates N value.

5.4 Discussion

These results are the first report of the potential of using honeybees as a component to improve greenhouse tomato yields in Kenya. They demonstrated that poor fruit set and yield of tomato under greenhouses may be due to pollination limitation, and honeybees (*Apis mellifera*) or manual truss shaking could be used to improve fruit set, size, and weight of the fruits. Correlation results showed the higher the number of seeds per fruit, the larger and heavier the fruit. This indicates

the additional yield benefits farmers are likely to reap if they considered supplemental pollination of greenhouse tomatoes. *M. bocandei* and *M. ferruginea* did not visit tomato flowers and were therefore considered not important in the pollination of tomato under enclosed environments. They were unable to forage for pollen from these flowers and hardly accepted the supplemental nectar in the form of honey-water offered to them. Despite the robust size of *M. bocandei*, it is very sensitive and cannot survive under enclosed environments such as greenhouses. It is an endemic stingless bee species found in Kakamega Forest and nests only on mature forests (Gikungu, 2006). The movements of the hives from Kakamega region to Nairobi area and the glasshouse environments could have affected the colony growth and development (Gikungu, pers. comm.). However, *M. ferruginea* was considered versatile as far as acclimatization to enclosed environments is concerned. Stingless bees play important ecological role as pollinators of many wild and crop plants and offer feasible alternatives for crop pollination (Heard, 1999; Slaa *et al.*, 2006: Cortopassi-Laurino *et al.*, 2006). Considering the global decline of both wild and managed pollinators, further evaluations of their potential for greenhouse pollination of other crops such as courgettes, watermelon, strawberries, vegetable seed of crops such as *Brassica* spp., onions, and other ornamentals will be of great interest.

There were differences in the bee foraging behaviour of A. mellifera under different foraging space and greenhouse temperatures. They were erratic during the first few days after introduction especially in the small glasshouse (3 m x 4 m) at KARI-NARL site compared to the bigger greenhouse (30 m x 8 m) at the commercial site, Rongai. One explanation for the erratic behaviour could be attributed to the limited flight space and that only a few days (3 days) were allowed for acclimatization before the start of data collection begun on colony traffic and foraging behaviour. At the commercial site, foraging was mostly concentrated within first 10 m from the hive location and no foragers were observed at the furthest end of the 30 m long greenhouse. Higo et al. (2004) found that more honeybees were recorded within the hive location than the other parts of the greenhouse (> 2 ha) and concluded that under enclosed environments, their foraging range was reduced to about 100 m or less. In this study, the greenhouse at the commercial site, Rongai was smaller than those used by Higo et al. (2004). Perhaps, use of more evenly spread hives per ha could ensure more even distribution of foragers in the greenhouse. Higo et al. (2004) suggested 2-4 hive per ha evenly distributed throughout the greenhouse. These may apply to supplementary pollination in temperate climate especially during winter and may not be applicable to tropical climates like Kenya. Studies on stoking rates for honeybees for managed pollination under greenhouses are therefore necessary and are recommended. Environmental factors such as temperature and humidity are likely to

influence the foraging behaviour of bees under enclosures. However, there was only a slight effect on the colony traffic at the commercial site, Rongai. Other factor such as the distinct tomato floral volatiles may have repelled the bees. A study by Morse (2009) indicated that β -phellandrene, 2carene, α -pinene, and p-cymene compounds emitted by greenhouse tomatoes flowers (Bigdena and Clarance), repelled bumblebees, and reduced their pollination efficiency. Though such volatile composition is still unknown for Chonto FI and Plumpty F1 varieties, such floral scent could affect the attractiveness of tomato flowers to bees especially the smaller and scent sensitive stingless bees like *M. bocandei* and *M. ferruginea*.

One key question on the potential pollination of tomato flowers by honeybees is effective pollen removal from the poricidal anthers. Despite the fact that they are unable to buzz pollinate, during a visit to tomato flowers, they were observed to vigorously strike the anthers using their forelegs. A cloud of released pollen was observed as a result of this behaviour, and they were observed packing the pollen into their corbiculae. Cane et al. (1993) and Thorp (2000) referred to this behaviour as "drumming" or "milking" of poricidal anthers to release pollen. Noteworthy is the long time foragers on tomato flowers. Probably, this process of pollen collection took longer time as would be compared to a buzz pollinator like bumblebees. Results from this study indicates that supplemental pollination by honeybees would improve fruit set, size and weight compared to no facilitated pollination, and corroborates other results that they can be utilised to improve seed set, size and quality in tomato greenhouse production systems (Banda and Paxton, 1991; Cribb et al., 1993; Sabara and Winston, 2003). Higo et al. (2004) found no increase in tomato sizes with addition of honeybees to the bumblebee forage force. Results from repeated honeybee visits (>1) indicated no additional benefits when compared to single visits. It is possible that, once sufficient pollen is released subsequent flower visitations/vibrations do no result into additional pollination and better fruit yield. Morandin et al. (2001b) reported no additional yield benefits in tomatoes produced from heavily pollinated tomato flowers that received >2 bumble bee visits. Also, Hogendoorn et al. (2006) reported no yield differences in tomatoes that were buzzed more than two times by Amegilla chlorocyanea. Correlation results showed that yield parameters such as fruit weight, length, and maximum diameter increased as the number of seeds per fruit increased. Other studies have reported that the number of seeds per tomato fruit is directly affected by the amount of viable pollen deposited on the stigma (Pressman et al., 1998; Morandin et al., 2001a). It is therefore correct to state that facilitated pollination by either manual truss shaking or by honeybees increased the number of viable pollen deposited on the stigma, and translated directly to higher seed number and consequently

bigger and heavier fruits. But, the low fruit yield results from the experimental KARI-NARL site could be attributed to other environmental factors such as the low temperature or high humidity recorded in the glasshouse. Picken (1984), Rylski and Aloni (1994) found that low temperature, irradiation and high humidity resulted in improper ovary development, malformed flowers and the production of unviable pollen. Perhaps, the high humidity coupled with the low temperatures led to poor dehiscing of pollen from the flowers. It is not surprising that despite the manual truss shaking and honeybee visitations, tomato fruit yield parameters were not significantly different from no facilitated pollination at this site. These aspects are critical to the overall productivity of greenhouse tomato systems and must be considered during the construction of the greenhouse and the crop management procedures like irrigation/watering. Limited nectar and pollen resources and the enclosed conditions, may have negatively affected the colony foragers' activity and perhaps, colony growth. Factors that would have resulted in reduced colony growth include lack of nectar and limited pollen sources. Although, nectar may have been compensated for by the supplemental honey-water, limited pollen access only to tomato flowers may be one of factor contributing to the reduction of colony activity. Sabara and Winston (2003) noted that insufficient protein supply, limited only to tomato flowers reduced the colony activity and brood development.

Although the study shows that honeybees (*Apis mellifera*) can be utilized by farmers to achieve better tomato pollination and yield in greenhouses, information on their management under these conditions for satisfactory crop pollination is still lacking. For example, due to honeybee erratic behaviour especially under small sized enclosures, care should be taken when handling them under enclosed conditions. Leaving vents open, would allow unlimited forage by the colony, as adult foragers are able to fly out and access additional pollen from other co-flowering plants near the greenhouse. But, co-flowering plants have been indicated to reduce pollination efficiency of honeybees (Cribb *et al.*, 1993; Sabara *et al.*, 2004). Hives could be replaced with new hives after every 3 weeks. To minimize loss of foragers and the possibility of greenhouse workers attack, introductions and removals of honeybee colonies should be undertaken late in the evening probably at dusk. Higo *et al.* (2004) suggested a one-week acclimatization period after introduction before greenhouse workers could resume normal crop maintenance work. Considerations should also be taken to ensure no pollination gaps exits between introductions and removals of colonies.

5.5 Conclusions

Most the farmers either selling their tomato produce in terms of weights or in heaps of different sizes. Investment in supplemental pollination would lead to an improvement in the two yield/quality parameters with possible economic benefits to the farmers. Considering the practicability of a choice of pollinating agent however, manual truss shaking has potential to increase fruit yield but is labour-intensive and expensive (Cribb et al., 1993; Ilbi et al., 1994). Moreover, the strength of shaking could differ relative to the person carrying it out and could result into damaged or premature loss of flowers. Use of honeybees (Apis mellifera) is a feasible alternative that farmers could utilize to achieve better tomato pollination and yield in greenhouses with minimal additional costs. Nonetheless, the increasing greenhouse production of crops presents a growing demand of pollinators suited for such enclosed conditions. Introductions of exotic bumblebees are likely to present environmental problems through forage competition with native bees, proliferation of weeds and alien plant species as indicated by a study in New Zealand (Hanley and Goulson, 2003) and should not be encouraged. Although in this study honeybees gave promising indication of their potential to improve fruit set and yield of tomatoes, more research should target other buzz pollinating bees such as Amegilla and Xylocopa. These bees have been successfully evaluated in other countries like Australia for pollination of tomatoes under cages. Hogendoorn et al. (2000) found that tomatoes pollinated by two Australian carpenter bees inside a flight cage, produced heavier fruits with higher number of seeds thatn those produced outside the flight cage. Kasina (2007) recorded X. calens and Halictus spp., visiting tomato flowers in the farmlands around Kakamega Forest, Western Kenya. Breeding protocols and evaluations of these species for pollination of greenhouse tomatoes will be of high interest.

6. QUANTIFYING THE ECONOMIC VALUE OF INSECT POLLINATION SERVICE TO AGRICULTURAL PRODUCTION IN KENYA

6.1 Introduction

Insect pollination is essential in the reproduction of many plants, and is a critical ecosystem service important for maintaining healthy and functional environments (Kearns *et al.*, 1998). An ecosystem service is defined by De Grout (1992) as "the capacity of the ecosystem to provide goods and services that satisfy human needs, directly or indirectly". It is estimated that 87 out of the 123 cultivated crops relies on insect pollination for fruit and seed set (Klein *et al.*, 2007). Roubik (1995) showed yield increment in about 70% of the tropical crops. Despite its critical role, insect pollination in agricultural production is still not recognized especially in developing countries. In Kenya, for example, pollination service is synonymous to honeybees and beekeeping products (honey, beewax), but ignores the contribution of pollinators to food crops (fruits, nuts, vegetables, oils), and other animal products derived from insect-pollinated forage, and feeds products.

Studies at global scale have indicated overwhelming economic benefits from pollination service. Richards (1993) estimated the global pollination value to agriculture at US\$ 200 billion per year. Costanza *et al.* (1997) on the other hand provided an estimate of US\$ 120 billion per year while Pimentel *et al.* (1997) estimated the value pollination service for the same year at US\$ 200 billion. Recently Gallai *et al.* (2009) estimated the global value of pollination service at US\$ 200 (\in 153) billion per year. Several studies have been undertaken at national levels to evaluate the monetary contribution by pollinators to agricultural production in the various countries and regions. These include for USA (Levin, 1984; Southwick and Southwick, 1992; Morse and Calderone, 2000; Losey and Vaughan, 2006), United Kingdom (Carreck and Williams, 1998), and Australia (Gordon and Davies, 2003). Such economic estimates of pollination service at national level are lacking especially in the developing countries. Ricketts *et al.* (2004) estimated that pollination services from tropical forests increased coffee yields in Costa Rica by 20% translating to approximately US\$ 62,000 for two forest fragments. A recent study in Uganda found that pollination services by bees contributed approximately US\$ 150 million (62%) to coffee production, equivalent to 24% of annual earnings

from export of agricultural products and 3% of Uganda's GDP (Munyuli, 2010). Kasina *et al.* (2009) estimated that pollination services contributed approximately US\$ 3.2 million in Kakamega District, Kenya. These estimates were limited to only eight vegetable crops grown but would be higher if more crops were included. Nevertheless, these estimates play a crucial role of highlighting the importance of insect pollinators to agricultural production, food, and nutrition security, and justify for greater investment in research, policy formulations, and conservation of pollinators by governments and research institutions.

In the recent years, there have been reports of declining pollinators (Buchmann and Nabhan, 1996; Allen-Wardell et al., 1998; Steffan-Dewenter et al., 2005), but increasing crop dependence on pollinators (Aizen et al., 2008). Forest fragmentation, land clearing for agriculture and other developments, pesticides use, pest and diseases are some of the driving factors associated with this decline (Allen-Wardell et al., 1998: Kevan and Phillips, 2001; Biesmeijer et al., 2006). Effects of climate change may accelerate this decline trend even further (Memmott et al., 2007; Hegland et al., 2009; Kjøhl, 2011). However, the global demand for pollination service is on the increase as more efforts are made towards sustaintaining food production (Aizen et al., 2008; Aizen and Harder, 2009). According to Roubik (1995), about 1,330 plant species cultivated in tropics, 70% show high insect crop dependence for pollination, and perhaps this is likely to be even more in the event of pollinator decline or absence. With the recent reports of varroa mite (Varroa destructor, Acari: Varroidae) in East Africa (Faizer et al., 2009), and the lack of understanding of pollinators' economic worth that has led to minimal investments into pollinator protection policies, food production, income from beekeeping and the functioning of the ecosystem are at risk. Besides, pollination service is regarded as free service, lack of conservation initiatives at individual and national levels are the norm (Kasina et al., 2009). For example in Uganda, despite the high contribution to coffee export incomes, farmers were not willing to manage their lands to protect pollination services (Munyuli, 2010). Monetary value of pollination service to agricultural production may broaden the general public awareness on the importance of pollination service and warrant some consideration by farmers, policy makers, and agricultural research scientists in relation to research and conservation.

So far, several approaches have been employed in estimating the monetary value of pollination services to agricultural production. A number of reviews of these approaches have also

come up as well (Mburu *et al.*, 2006; Hein, 2009). According to Mburu *et al.*, (2006) these approaches include:

- (i) Contingent valuation method (CVM) approach utilises the willingness to pay (WTP) for a theoretical pollinator protection policy to estimate the total value of pollination service. Questionnaires are usually administered to a representative population seeking to value preferences of the public for a theoretical policy that would maintain and protect pollinators. The population interviewed must be fully aware of the benefits of pollination. Both direct use and the non-use values of pollination service are measured. Kasina (2007) used CVM and found that each household in Kakamega were willing to pay about US\$ 90 annually to conserve pollinators.
- (ii) Replacement cost method uses the cost proxy approach to estimates the value of pollination service. For instance, the cost of replacing pollinators' e.g through hand pollination of vanilla or apples flowers in absence of pollinators. Allsopp *et al.* (2008) used this approach to estimate the value of PS for the Western Cape deciduous fruit industry of South Africa.
- (iii) Production function (Bio-economic) method considers PS as a direct input to crop production along other inputs such as fertilizers and other fixed inputs used in the production of agricultural output. The effect of pollinators varies from crop to crop ranging between 0% to over 90% (Klein *et al.*, 2007). This contribution can be directly equated with its impact on the production of the marketed output (Mburu *et al.*, 2009). Carreck and Williams, 1998: Gallai *et al.*, 2009; Kasina *et al.*, 2009 amongst other authors have used this approach. Mburu *et al.* (2006) recommended this method for estimations for the national aggregate value of pollination service. A recent report published by FAO gives guidelines to be used while using this methodology to assess the value of pollinaton service at national level and vulnerabilities to pollinator declines (Gallai and Vaissière, 2009).

In the current study, bio-economic approach was used to estimate the value of pollination service to Kenya's agricultural production. One major challenge faced with this method was the unavailability of crop's insect dependency ratios. Unlike in developed countries like Europe, USA, and Canada, where insect pollinator dependence ratios are known for most crops, very few pollination studies have been undertaken in developing countries like Kenya (Rodger *et al.*, 2004).

Available studies are on watermelon (Njoroge *et al.*, 2004), bottle gourd/dudhi (Morimoto *et al.*, 2004), tomatoes, capsicums, passion fruits, pumpkins (Kasina, 2007), eggplant (Gemill-Herren and Ochien'g, 2008), sunflower (Nderitu *et al.*, 2008), and papaya (Martins and Johnson, 2009). Pollinator dependency ratios for most crops considered in this analysis were derived from Klein *et al.* (2007), Gallai *et al.* (2009) and Kasina *et al.*, 2009. In addition, pollinator dependence values established in chapters three, and four of this current study were used. The study assumed that, although crop pollination requirements and pollinator diversity may differ depending on location as well as crop varieties, the differences might not be significant to change the interpretation of the results. In any case, the crop pollination dependency ratios by Klein *et al.* (2007) have been used recently in evaluation of the global economic value of pollinators-mediated crops to the nutrients in the human food supply. For this study, additional pollinator dependence ratio established for AILVs and other crops in chapters three and four were used. Nevertheless, these estimates are expected to be informative and provide baseline data on the economic contribution of insect-pollinators to agricultural production in Kenya.

6.2 Materials and methods

6.2.1 Economic value of pollination service estimation approach

In this study, attempts were made to estimate the value of pollination service for selected crops grown in Kenya using the bio-economic approach. It uses the crops dependency ratios on pollinators for yield and quality to value pollination service to agricultural production. This approach has been used by many authors (Robinson *et al.*, 1989; Southwick and Southwick, 1992; Morse and Calderone, 2000; Losey and Vaughan, 2006; Kasina *et al.*, 2009) and recently by Gallai *et al.* (2009) to calculate the world's agricultural vulnerability in case of pollinator loss.

Total annual economic value of the crop $(EVi) = (Q_i \times P_i)$ (1)

The total economic value of pollination service (IPEV) was calculated as:

$$IPEV = \sum_{i=1}^{l} (Qi \times Pi \times Di)$$
⁽²⁾

Further, ratio of vulnerability (RV) of the Kenyan agriculture to loss of pollinators was calculated as defined by Gallai *et al.* (2009) and provides a measure of the relative production loss assuming lack or loss of pollinators. This was represented as:

$$RV = \frac{IPEV}{EV} = \frac{\sum_{i=1}^{l} (Pi \times Qi \times Di)}{\sum_{i=1}^{l} (Pi \times Qi)} \times 100$$
(3)

where:

- $Q_i =>$ the annual quantity (MT) of crop *i* produced in Kenya (HCDA, 2009; MOA, 2010),
- $D_i =>$ the dependency ratio of the crop *i* on pollinators (Klein *et al.*, 2007),
- $P_i \Rightarrow$ the annual value (US\$) of crop *i* produced produced in Kenya (HCDA, 2009; MOA, 2010).
- I => crops considered, i = 1, ...I.

6.2.2 Study crops

Study crops were defined according to FAO: oil seed crops, fruits crops, legumes and pulses, nut crops, roots and tubers, spices, sugar crops, stimulant crops and vegetables. Production figures and value data for these crops were sourced from the latest annual economic review of agriculture for the year 2009 (HCDA, 2009; MOA, 2010). The national total area under production (ha), production (MT) and the total monetary values (US\$) were given for each crop (see Appendix 6.1). This report included seed production and fresh leaves market values of various African Indigenous Vegetables (AILVs) including spider plant (*Cleome gynandra* L.), slender plant (*Crotalaria brevidens* Benth) and broad-leafed African nightshade (*Solanum scabrum* Mill) were also included in this analysis. However, production data on other legume crops utilized for forage such as dolichos (*Dolichos lablab*), lupins (*Lupinus* sp.), desmodium (*Desmodium uncinatum*), lucerne (*Medicago sativa*) and agroforestry plant species such as *Sesbania sesban* are not available. Seed production of these crops may benefit from pollinators.

Other than the estimations on the value of pollination at national level, a further analysis was performed at provincial level following the political boundaries of Kenya (eight provinces). According to the Agroecological Zones (AEZ) of Kenya, these regions have different agricultural potential depending on the amount of rainfall and other climatic conditions like temperature; crop diversity is distinct in some cases (Jaetzold and Schmidt, 1982). For example, cashew nut is grown within the Coastal province. It is speculated that, their vulnerability to pollinator loss will be variable. In addition, some pollinator species may be found only in some regions, and not others. Therefore, such a focus on the provinces will highlight most vulnerable agricultural production areas for future specific conservation strategies.

6.3 Results

Current estimates indicated that about 100 plant species are important agricultural crops in Kenya (Appendix 6.1). Over 40% of these crops are dependent on insect pollinators for fruit set and yield (Appendix 6.2). For 22% crops, pollinators are essential. About 2 million hectares (more than half of total national area under cultivation) are covered with pollinator-dependent crops. Total national value of agricultural produce for 2009 stood at US\$ 4 billion while the total national value for pollinator-dependent crops was US\$ 1 billion (Appendix 6.1). This indicates that 25% of the total national agricultural production value relies on insect pollinators. This is likely to increase in the future due to introduction of new crops and more intensive agricultural production geared to meet the food requirements of the growing population. Grain cereals took 50% of the total national area under crop production. They do not require insect pollination. The total value of pollination service to Kenya's agricultural production in 2009 was estimated to be US\$ 400 million (Table 6.1). This represented 1% of the total horticultural export for the same year. This economic value of pollination service was more than 6 times the total national annual value of bee products (US\$ 60 million).

When different crop categories were compared at national level, grain legumes had the highest economic benefits from pollination service, followed by fruits, vegetables, stimulant crops (coffee), nuts and finally AILVs seed. The value of pollination service to seed production for the three AILVs (spider plant, broad-leafed African nightshade and slender leaf) was estimated to be US\$ 313,000 representing 3% of their fresh leaf total value of US\$ 10 million for 2009.

Crop category	Total area	Total production	$EV (10^6 US\$)$	IPEV $(10^6 \text{ US}\$)$	RV (%)
	(10^3Ha)	(10^3MT)			
Grain legumes	1,198	614.3	430.09	149.51	34.8
Fruits	141.3	2,824	435.9 103.85		23.8
Vegetables	96.7	1,986.4	484.61	67.97	14
Stimulants (Coffee)	160	47.8	138.96	34.74	25
Nuts	51.2	60.7	21.65	7.9	36.5
Oil crops	40.0	14.89	5.03	5.03 1.26	
AILVs seed	*	0.01	0.39 0.31		79.5
Grain cereals ^a	2,319.9	2,871.8	1,117.55	0	0
AILVs (leaves) ^a	27.3	157.7	33.96	0	0
Sugar cane ^a	154.3	548.2	557.61	0	0
Spices and Herbs ^b	1.1	5.5	21.05	21.05 0	
Pyrethrin extract ^b	4.1	0.01	12.32	12.32 0	
Root crops ^a	259.9	4,524.7	686.68	0	0
Totals	4,611.5	13,970.2	3,968.9	365.54	9.2

Table 6.1: Economic value of pollination service (IPEV) to Kenya's agricultural production and vulnerability ratios (RV %) to pollinator loss, 2009

^a Yield not pollinator-dependent and ^b yield pollinator-dependent but no reported pollinator dependence ratio and ^ccottonseed only,* total area under production not available, AILVs (African Indigenous Leafy Vegetables). Pollination dependence values adapted from Klein *et al.* (2007), Gallai *et al.* (2009), Kasina *et al.* (2009) and chapters three and four of this thesis. National production and average prices adopted from HCDA (2009) and MOA (2010) Exchange rates US\$ 1 = 77.0 (CBK, 2009).

The vulnerability ratio of national agricultural sector to insect pollinator loss was 9% but increased to 35% when only pollinator-dependent crops were considered. AILVs seed production was the most vulnerable (almost 80%) followed by nuts (37%), grain legumes (35%), stimulants (coffee) (25%) and fruits (24%) (Table 6.1). At the individual crop level, common beans had the highest monetary gain from insect pollination service US\$ 114 million, mangoes at US\$ 53 million, tomatoes at US\$ 52 million and coffee (US\$ 35 million) (Table 6.2).

Crop catergory	Pollinator dependence ratio	Total revenue (EV) US\$ 10 ⁶	IPEV (EV) US\$ 10⁶
Grain legumes			
Common beans	0.33	344.76	113.77
Cowpeas	0.41	47.77	19.58
Greengrams	0.43	37.56	16.15
Fruits			
Mangoes	0.65	81.65	53.07
Papaya	0.95	18.64	17.71
Avocado	0.65	18.82	12.23
Watermelon,sweet and muskmelon	0.95	12.29	11.67
Passion fruit	0.28	16.96	4.75
Citrus	0.05	35.47	1.77
Apples	0.65	0.91	0.59
Plums	0.65	0.68	0.44
Pears	0.65	0.54	0.35
Guavas	0.65	0.52	0.34
Loquats	0.60	0.24	0.14
Custard apples	0.95	0.11	0.10
Peaches	0.65	0.08	0.05
Strawberries	0.25	0.05	0.01
Nuts			
Cashew nuts	0.65	6.35	4.13
Macadamia nuts	0.65	5.01	3.26
Groundnuts	0.05	10.29	0.51

Table 6.2: List of individual insect-dependent crops cultivated in Kenya, total revenue, pollinator dependence ratios and estimated economic value of insect pollinators, 2009

Crop catergory	Pollinator dependence ratio	Total revenue (EV) US\$ 10 ⁶	IPEV (EV) US\$ 10⁶	
Vegetables				
Tomatoes	0.25	207.21	51.80	
Pumpkins	0.95	6.27	5.95	
Capsicums	0.66	5.22	3.45	
Courgettes	0.95	2.77	2.64	
Butternuts	0.95	2.33	2.22	
French beans	0.05	22.58	1.13	
Cucumber	0.65	0.92	0.60	
Dudhi/bottle gourd	0.95	0.44	0.42	
Karela	0.95	0.18	0.17	
Chillies	0.05	2.75	0.14	
Okra	0.1	1.29	0.13	
Turia	0.95	0.13	0.12	
AILVs seeds				
Spider plant	0.99	0.21	0.21	
Slender leaf	0.98	0.09	0.08	
Broad-leafed African nightshade	0.1	0.08	0.01	
Stimulants				
Coffee	0.25	138.96	34.74	
Oil crops				
Cottonseed	0.25	5.03	1.26	
Totals		1,035.16	365.73	
Vulnerability ratio			35.3	

Table 6.2: Cont. List of individual insect-dependent crops cultivated in Kenya, total revenue, pollinator dependence ratios and economic value of pollination service, 2009

Pollination dependence ratios adapted from Klein *et al.* (2007), Gallai *et al.* (2009), Kasina *et al.* (2009) and chapters three and four of this thesis. AILVs (African Indigenous Leafy Vegetables). National production and average prices adopted from HCDA (2009), and MOA (2010). Exchange rates US\$ 1 = 77.0 (CBK, 2009).

Further results showed variable results across the eight provinces in Kenya. Eastern province had the highest economic (US\$ 74 million) gain from pollination service followed by Nyanza (US\$ 61 million) and Coastal province (US\$ 48 million). Depending on the type of crops grown, production, and its dependence on pollinators fruit/yield, the economic vulnerability ratios varied between 5% to over 50% among the provinces (Table 6.3, Appendix 6.2).

Crop category/ Province		Central	Coast	Eastern	Western	Nyanza	Rift Valley	Nairobi	North Eastern
Grain legumes ¹	Production (10^3 MT)	55.1	1.4	172.6	63.9	136.2	992.9	0.2	0.013
	EV(10 ⁶ US\$)	30.05	1.03	127.91	47.62	106.60	68.97	0.11	0.1
	IPEV (10 ⁶ US\$)	9.92	0.35	42.24	15.88	38.63	22.86	0.04	0.003
	RV (%)	33	34.1	33	33	36.2	33.1	33	33
Fruits	Production (10^3 MT)	35.3	466.1	181.8	20.3	84.6	76.8	5.8	4.5
	EV(10 ⁶ US\$)	11.69	82.22	40.79	6.94	26.82	17.48	1.41	1.57
	IPEV (10 ⁶ US\$)	7.44	38.79	23.87	4.52	4.35	9.23	0.92	1.45
	RV (%)	63.6	47.2	58.5	65.1	16.2	52.8	64.8	91.9
Vegetables ²	Production (10^3 MT)	173.6	52.4	75.3	43.0	170.6	91.6	3.5	18.7
	EV(10 ⁶ US\$)	81.42	17.03	24.96	21.34	65.85	28.58	0.82	5.88
	IPEV (10 ⁶ US\$)	18.91	4.34	5.50	5.45	17.11	7.39	0.26	1.55
	RV (%)	23.3	25.5	22	25.5	26	25.9	31.5	26.3
Nuts	Production (10^3 MT)	5.3	25.2	10.4	0.5	18.0	1.4	0	0
	EV(10 ⁶ US\$)	1.23	6.70	4.22	0.26	8.18	1.06	0	0
	IPEV (10 ⁶ US\$)	0.80	4.35	2.14	0.02	0.41	0.20	0	0
	RV (%)	65	65	50.7	5.9	5	17.1	0	0
Total	EV(10 ⁶ US\$)	124.39	106.98	197.88	76.17	207.45	116.09	2.35	7.46
	IPEV (10 ⁶ US\$)	37.01	47.83	73.77	25.87	60.50	39.67	1.21	3.0
	RV (%)	29.8	44.7	37.3	34.0	29.2	34.2	51.6	40.2

Table 6.3: Economic value of pollination service (IPEV) to agricultural production and vulnerability ratios (RV%) to pollinator loss, at provincial level in Kenya 2009

¹ Grain legumesincluded only beans, cowpeas and greengrams. ²pumpkins, cucumbers, courgette/squash production data excluded from vegetables. Stimulants (coffee) and oil crops data excluded. Pollination dependence values adapted from Klein *et al.* (2007), Gallai *et al.* (2009), Kasina *et al.* (2009) and chapters three and four of this thesis. National production and average prices adopted from MOA (2010), and HCDA (2009). Exchange rates US\$ 1 = 77.0 (CBK, 2009).

Generally, three provinces were most vulnerable to pollinator loss were Nairobi (52%), Coast (45%), and North Eastern (40%) (Table 6.3). For some crop categories, e.g fruits and nuts, higher vulnerability ratios were recorded across the provinces compared to the national aggregate vulnerability ratio.

6.4 Discussion

The economic value of pollination service to agricultural production was estimated at US\$ 400 million. This is the direct value of pollination service using the bio-economic (production function) approach. It measures production for most crops that is directly associated with insect pollinators. Perhaps higher figures would be realised with other methods of valuations such as replacement cost approach and contingent valuation method (CVM). For instance, CVM utilises the willingness to pay a theoretical pollinator conservation policy to estimate the value of pollination service and would include the non-use value of pollination service. Non-use value in pollination context has been characterised by Kolstad (2000) as existence value (utility derived from insect pollinator existence), altruistic value (utility derived from the benefits other people enjoy from insect pollinators), and bequest value (utility from pollinators that the future generations would enjoy from insect pollinators). Also, the insect crop dependence ratios used in this study may be different (higher or lower) in space and time. However, changes in these ratios are not expected to significantly affect the resultant estimates in a negative manner. In fact, the estimated value of pollination service may increase substantially if the role of pollinators in seed production for flowering plants used in home gardens, leafy vegetables, herbs, spices, and fodder crops were considered. Due to unavailability of data, they were not included in the current analysis. Fodder crops, for example, desmodium (Desmodium uncinatum, D. intortum: Fabaceae), lucerne (Medicago sativa: Fabaceae), dolichos (Dolichos lablab), lupins (Lupinus albus and L. angustifolus: Fabaceae) are utilised to feed animals which in turn produce important products and by-products i.e milk, meat, butter and cheese etc. Others where pollinator could be preponderant include seed production of agro-forestry plant species e.g sesbania (Sebania sesban and S. grandiflora: Fabaceae) and the reproduction and regeneration of wild flora and fauna that maintain the general ecosystem functions in the environment. Nevertheless, these estimates will increase the public awareness of the importance of pollination service to agricultural production in Kenya.
During the year under consideration, about 2 million hectares of land were grown with pollinator-dependent crops. Introduction of new crops like vanilla (*Vanilla planifolia*), moringa (*Moringa oleifera*) and jatropha (*Jatropha curcus*) is likely to change the scenario. Adoption of new farming practises such as greenhouse cultivation of vegetables e.g capsicum and tomatoes will require that farmers invest in managed crop pollination to increase yield and quality. Under greenhouse conditions, tomato dependence on insect pollinators increases from little (0.25) to great (0.65) (Vaisiére pers. comm.). But, the upcoming vanilla production would rely exclusively on hand pollination as its natural pollinator; the stingless bee *Melipona*, is absent in Kenya. It is clear the above trend point towards increasing demand for pollinator-dependent crops especially in developing countries. Given such a scenario, conservation of pollinators is very essential to both sustainable agricultural production and healthy ecosystem function.

The low vulnerability to pollinator loss (9%) reported in this study may misguide one to conclude that the loss of insect pollinators would not present a threat to the GDP and food security. On the other hand, it presents very high threats to the nutritional balance and security. Main food crops such as cereals and root crops do not require insect pollination. Vital vitamins, antioxidants and other nutritional elements are mainly obtained through consumption of fresh fruits and vegetables. Recently, Eilers *et al.* (2011) found that more than 90% of Vitamin C, majority of Lycopene and β -cryptoxanthin and β -tocopherol oxidants, lipids, vitamin A and related carotenoids, calcium and fluoride and large proportions of folic acid depend fully or partially on pollinators and that more than 40% of some essential nutrients provided by fruits and vegetables could be lost without pollinators. With the global pollinator decline, this may worsen the effects of malnutrition especially in developing countries even further. High vulnerability ratios of over 90% were recorded for important fruits and vegetables within the cucurbitaceous family. These crops are monoecious (female and male flower parts on the same plant but at different locations within the plant), the pollen is heavy and sticky and the flowers are opened only for a brief period of time (Free, 1993). Pollinators are therefore very essential if any yields are to be realised or improved.

Regional differences in terms of vulnerability to pollinator loss could be attributed to the uniqueness of these regions in terms of crop types cultivated across the regions. These results demonstrated that agricultural production in Eastern and Coastal province were more pollinator dependent compared to other ares e.g Nyanza and Central provinces. This is because certain crops

such as cashew nuts are only cultivated at the Coastal region while macadamia nut is grown in Eastern province. These crops have high insect pollinator dependence (0.65) (Klein et al., 2007). Despite the economic contribution of macadamia and cashew nuts to agriculture and the fact that higher yields may be realised with better pollination, no records of their pollinators exists in Kenya. In the event of pollinator loss, national revenue of about US\$ 8 million annually would be lost. Considering Nairobi and North Eastern provinces, the vulnerability to pollinator loss could be even higher in the near future due to the increasing encroachment of the urban settlement areas which are important pollinator refuge areas and the introduction irrigation agriculture respectively. In the latter case, as more land is being converted to meet the increasing food demand, pressure on the biodiversity is likely to increase, while at the same time the demand for pollination service for agricultural production is expected to increase. This could be catastrophic given that some of these regions could harbour unique pollinator species, and perhaps endemic bee species e.g stingless bee Meliponula bocandei may be endemic to Kakamega region (Gikungu pers. comm.). Apparently, economic gain from pollination service in Western province was high (US\$ 26 million) as compared to Kasina et al. (2009) of US\$ 3.2 million for Kakamega district in 2005. While Kasina et al. (2009) considered only limited number of crops (eight crops) produced by smallscale farmers within Kakamega district, the current estimates encompassed many crops (over 30) within the larger Western provincial area.

6.5 Conclusions

One limitation of this study was lack of information on pollinator dependence ratios and production data for important crops e.g pyrethrum, coconut, sunflower seed, pigeon peas, fruits (white sapota, turia), and vegetables. It is recommended, that urgent crop pollination studies be carried out to gain more precise pollination needs and effective pollinators of most crops. Spatial distribution of the pollinators together with important floral plants will be crucial in sustainable pollinator management and conservation strategies. Limitations notwithstanding, the estimates in this study indicate that pollination service is crucial to Kenya's agricultural production and that any loss of pollinators may significantly affect agricultural production, food, and nutrition security. The annual economic value of pollination service is many more times higher than the annual value of honeybee products (honey and beewax). These results will be indispensible to decision/policy makers for effective argument for conservation policies and designing cost effective sustainable pollinator

diversity conservation programmes. In fact, such conservation measures are urgently needed, if any considerable sustainable agricultural production is to be achieved. On the other hand, other than improved awareness on the relative benefits of pollination services and pollinator friendly ecological agricultural practises, these estimates may provide them with incentives to conserve their habitats as well.

7. GENERAL CONCLUSIONS AND RECOMMENDATIONS

The overall objective of this study was to contribute to the knowledge of pollination and pollinators of agricultural crops in Kenya. The specific objectives were (1) describe the pollination needs and seed quality of African Indigenous Leafy Vegetables (AILVs), (2) assess the diversity and effectiveness of pollinators of selected vegetable crops grown around Kakamega Forest, Western Kenya (3) to evaluate the potential of honeybees for the pollination of greenhouse tomatoes in Kenya, and (4) undertake an economic analysis of the contribution of pollination service to agricultural production in Kenya.

7.1 Major findings

Results indicated that African Indigenous Leafy Vegetables require pollinators for seed set and quality (Chapter 3). Other than the broad-leafed African nightshade (*Solanum scabrum*), spider plant (*Cleome gynandra*) and slender leaf (*Crotalaria brevidens*) rely heavily on pollinators for seed set (over 90%). Their seed set and quality were considerably reduced when pollinators were excluded. Spider plant is nocturnally receptive and is pollinated by sphingid hawkmoths.

Contrary to the belief that all floral visitors are synonymous to pollination success, the results showed that crops may only be effectively and persistently pollinated by distinct pollinators despite a wide range of floral visitors. Using Spear's (1983) index of single visit pollinator efficiency, the direct measure of the floral visitor to seed set was evaluated. An in-depth knowledge of the pollinating agent is necessary for managed pollination. Measured by their effectivess on the seed set for various vegetable crops including three African Indigenous Vegetables (AILVs) (Chapter 4), hawkmoth species, *Hippotion osiris, H. eson, Agrius convolvuli* and *Nephele aequivalens* were considered important pollinators for spider plant (*Cleome gynandra*). *Megachile* sp., and *Xylocopa* sp. were the most effective pollinators for slender leaf (*Crotalaria brevidens*). Although broad-leafed African nightshade (*Solanu scabrum*) is wind pollinated and self-fertile, visits by *Amegilla langi* and *Xylocopa calens* increased seed set. *Lasioglossum* sp., *Apis mellifera, Pleibena hildebrandti* effectively pollinated karela (*Momordica charantia*). *Xylocopa inconstans*, and *X. calens* were the

most important bee pollinators for eggplant (*Solanum melongena*). *Lasioglossum* sp. showed high potential for the pollination of okra (*Abelmoschus esculentus*).

Use of honeybees (*Apis mellifera*) for pollination of greenhouse tomatoes resulted into improved fruit set, and yield (Chapter 5). Fruits from flowers visited the bees and those that received manual truss shaking treatment were bigger and weighed heavier compared to no facilitated pollination. These results show that farmers may introduce *Apis mellifera* hives into greenhouse with flowering tomato. Alternatively, hand tappings of the flower clusters to release pollen, may increase fruit set and yield. However, more education for farmers on the management practises is necessary in order to optimise pollination benefits form honeybees.

Using the bio-economic approach, an attempt to quantify the national economic benefits derived from pollination service revealed that about US\$ 400 million of the total national value of agricultural production for the year 2009 was attributed to insect pollinators (Chapter 6). This presented about 9% vulnerability for the total national agricultural production incase of pollinator loss. However, the vulnerability ratio increased to 35% when only pollinator-dependent crops were considered. The most vulnerable were legumes, nuts, fruits, and vegetables indicating high risks of malnutrition when faced with such pollinator decline. The eight provinces showed variable economic benefits from pollination service as well as vulnerability to pollinator loss. Eastern province had the highest economic gain from pollination service (US\$ 74 million), Nyanza (US\$ 61 million) and Coastal province (US\$ 48 million) while Nairobi was the most vulnerable to pollinator loss (52%), Coast (45%), and North Eastern (40%).

7.2 General conclusions

The results in this study highlighted the importance of pollinators in the seed yield and quality of the selected African Indigenous Vegetables. Except for *Solanum scabrum*, pollinators are essential in the seed production of *Cleome gynandra* and *Crotalaria brevidens*. It is further recommended that for increased seed set as well for improved quality seed production sites of these AILVs should be located near natural habitat such as forest. More studies should be carried out in these commercial seed production areas e.g Bungoma, Kitale, and Thika, to determine the status of pollinators and the potential effect on seed set and quality. The increasing interest in AILVs vegetables and the fact that seed companies are interested in production of high quality seeds, signify

that AILVs are important emerging crops that would contribute significantly to the overall agricultural economic development. It is therefore important that more research be focused on the modes of enhancing pollinator population, for example through habitat rehabilitation, and reduced use of pesticides by embracing an integrated pest management approach for improved pollination service. The pollination requirements of other AILVs e.g jute mallow (*Corchorus* sp.), cowpeas (*Vigna unguiculata*), Kahurura/fig leaf gourd (*Cucurbita ficifolia*) are still lacking and should be undertaken.

These findings provide the first quantification of the pollinator effectiveness of floral visitors to crops and contributes to the growing literature demonstrating the importance of solitary bee species as well as other non bee pollinators in crop pollination. The results indicated the critical role played by pollinator on the seed set and quality of AILVs. Spider plant seems to be highly pollen limited. Possibly, seed producers may consider hand pollination if high quality seeds are to be made available to farmers. Other than being expensive in terms of labour costs, hand pollination must only be carried out at dusk thus complicating its practicability of its execution. Alternatively, management of hawkmoth habitats e.g through ruderal patches and diverse hedgerows may improve the diversity and abundance of Hippotion osiris, H. eson, Agrius convolvuli and Nephele aequivalens. Identification of larvae food preferences may be important in providing specific plants that could be targeted in such hedgerows restoration or improvement programmes. In addition, the utilization of non-Apis species as crop pollinators is emphasized as a viable alternative method of meeting future pollination needs in rapidly changing agricultural environments. Observation of Plebeina hildebrandti visiting flowers of M. charantia is in particular of much interest and highlights the potential of stingless bees for crop pollination of commercially important crops. Further research on the potential of stingless bees and solitary bees as manageable crop pollinators will be of interest. However, these results only presented data collected from a single locality and it would be expected that pollinator diversity, abundance and their effectiveness may be different at other localities from those observed here. More pollination studies in other locations are therefore recommended. Nevertheless, based on these findings that non-Apis pollinators are important in crop production, relevant pollinator conservation measures can be instituted as opposed to Apis-biased conservation strategies.

7.3 General recommendations

- 1) Most crop production in Kenya is carried out by the small-scale farmers who other than having a diverse range of crops in one farm, rely heavily on the unmanaged wild bee pollinators for crop pollination. There is need to identify among the native pollinators, potential species for managed pollination and examine the possibility of mass culture/breeding of them. For instance, *Megachile rufipes*, effective in the pollination of *Crotalaria brevidens*, are potential candidates for such venture of commercialised pollination. They are gregarious and readily accept artificial nests. Studies on their breeding biology are urgently required. The same would apply to *Lasioglossum* and *Xylocopa* species. Since there is no managed pollinator at the moment, farmers may adopt an ecosystem management approach to benefit from improved pollination by identifying crop pollinators and their corresponding habitat requirements e.g pollen and nectar sources. Such measures may be extremely useful in increasing the diversity and abundance of native pollinators, crop pollination, and agricultural production.
- 2) Maintaining natural patches within and surrounding farmlands would ensure availability of nectar and pollen during those periods when crops are not flowering. These areas would include diverse hedgerows, uncultivated patches within cropping area, and flower gardens in residential areas. Although this may imply a reduction in crop production area, it is likely that farmers will record higher yields due to increased pollinator diversity, abundance and the presence of other beneficial insects e.g insect pests, parasites and predators. However, the net economic gain for such agricultural landscape planning and habitat restoration initiatives should be investigated in order to provide economic argument and incentives to farmers for conservation and management of these natural habitats.
- 3) It is well established that improper use of agricultural pesticides adversely affects the development of honeybees. However, the effect on other wild bees and other non-bee pollinators remain unknown, and is critical for the protection of pollinator populations. In this study, hawkmoths were effective pollinators for *Cleome gynandra*, but their larval stages are considered important agricultural pests and their control would be catastrophic in terms of seed set and reproductive success of other wild plants that rely on them for pollination. In addition, accumulations wash-off pesticides into the soil may accelerate the decline of soil nesting bees, e.g *Lasioglossum*. Long-term studies on the cumulative effects of pesticide use on pollinators are

necessary. Efforts to mitigate these effects may include pollinator friendly integrated pest management (IPM) strategies, e.g using pesticides that are less toxic to pollinators, spraying at times when pollinators are not active. Rigorous testing experimentation/protocols on the effect of pesticides on bees should not only focus on honeybees, but also other wild bees in light with the significant agricultural pollination service they provide.

- 4) Nesting sites of these pollinators must be protected. While Lassioglossum sp., Pleibena hildebrandti, are soil nesters, Megachile sp. and Xylocopa sp. nests in dry wood and stems, other floral visitors important in the crop-pollinator web may nest in abandoned termite mounds, and other groundbreaking contructions may negatively influence their populations. Research focused on the development of protocols for management and mass culture of these species with focus for commercial manged crop pollination are urgently needed.
- 5) Meliponiculture provides new emerging frontiers for alternative pollinators for crop production and income generation for the farmers. Observation of *Plebeina hildebrandti* visiting flowers of *M. charantia* is in particular of much interest and highlights the potential of stingless bees for crop pollination of commercially important crops. Further research on the potential of stingless bees and solitary bees as manageable crop pollinators will be of interest.
- 6) Changing production system for example, greenhouse pollination of crops as well as introduction of new crops is likely to increase the demand for managed pollination but importation of exotic bees should not be encouraged as it is likely to cause disruptions of the local pollinator species. To meet this demand, evaluation of breeding, mass culture of other pollinators such as *Amegilla* and *Xylocopa* species for managed greenhouse crop pollination of e.g tomatoes, capsicums, watermelon, vanilla, is recommended.
- 7) Government involvement in pollination research of both crops and wild plants is urgently needed. Pollination service contributes significantly to the economic development, food, and nutritional security. Yet, minimal consideration by the government for example funding of research and conservation of pollinators are wanting. There is flagrant absence of information on the crop pollinators in Kenya. To fill these gaps, public awareness creation of a wide range of issues dealing with pollinators is urgently needed at both the pollitical, professional and individual farmer's level.

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9. APPENDICES

Appendix 6.1: Important agricultural crops in Kenya, total national area under cultivation, production and total revenue, 2009

No.	Crop categories	Area under production (Ha 10 ³)	Total Production (MT 10 ³)	Total value US\$ 10^6	
	Grain cereals				
1.	Maize (Zea mays)	1,885.1	2,442.82	921.43	
2.	Wheat (Triticum aestivum)	131.6	219.30	113.0	
3.	Rice (Oryza sativa)	21.8	42.2	*	
4.	Barley (Hordeum vulgare)	3.7	16.07	*	
5.	Sorghum (Sorghum vulgare)	173.2	94.96	45.01	
6.	Millet (<i>Pennisetum</i> galucum)/Finger millet (<i>Eleusine</i> coracana)	104.6	56.42	38.10	
	Grain legumes				
7.	Common (field) bean (<i>Vicia faba</i> var. minor)	960.7	465.36	344.76	
8.	Cowpeas (Vigna unguiculata)	124.3	60.15	47.77	
9.	Green grams (Vigna radiata)	113.0	42.33	37.56	
10.	Pigeon peas (Cajanus cajan)	118.2	46.47	*	
	Nuts and oil crops				
11.	Bambara nuts (Vigna subterranea	*			
12.	Groundnuts (Arachis hypogaea)	20.6	21.45	10.29	
13.	Macadamia nuts (<i>Macadamia intergrifolia</i>)	2.0	14.78	5.01	
14.	Coconut (Cocos nucifera)	*			
15.	Cashew nuts (Anacardium occidentale)	28.6	24.46	6.35	
16.	Soybean (Glycine max)	1.5	1.35	*	

No.	Crop categories	Area under production (Ha 10 ³)	Total Production (MT 10 ³)	Total value US\$ 10 ⁶
17	Sunflower (Helianthus annus)	*		
18.	Cotton seed (Gossypium spp.)	40.0	14.89	5.03
	Fruit crops			
19	Bananas (Musa spp.)	69.9	1,686.93	189.0
20.	Papaya (Carica papaya)	7.3	98.68	18.64
21.	Avocado (Persia americana)	4.2	70.81	18.82
22.	Mangoes (Mangifera indica)	32.7	474.61	81.65
23.	Guava (Psidium guajava)	1.1	6.01	0.52
24.	Citrus sp.(oranges, lemon, lime, tangerine, grapefruit)	11.4	138.83	35.47
25.	Passion fruits (Passiflora edulis)	3.2	39.8	16.96
26.	Pineapples (Ananas comosus)	7.9	257.62	57.75
27.	Watermelon (Citrullus lanatus	2.0	36.03	12.29
28.	Straw berries (Fragaria spp.)	0.01	0.05	0.05
29.	Apples (Malus domestica)	0.07	1.25	0.91
30.	Plums (Prunus spp.)	0.12	1.49	0.68
31	Pears (Pyrus communis)	0.2	2.44	0.54
32	Peaches (Prunus persica)	0.03	0.39	0.08
33.	Grapes (Vitis spp.)	0.11	0.37	0.25
34	Custard apple (Annona reticulata)	0.12	0.64	0.11
35	Loquats (Eriobotrya japonica)	0.21	0.001	0.24
36.	Tree tomato (Solanum betaceum)	0.4	2.86	0.85
37	White sapote (Casimiroa edulis)	0.07	0.74	0.18
	Vegetables			
38.	Cabbage (Brassica oleracea)	- 14.8	627.83	112.36
39.	Kales (Brassica sp.)	25.1	356.87	55.91

Appendix 6.1: Cont.	Important	agricultural	crops	in	Kenya,	total	national	area	under	cultivation,
production and total re-	evenue, 200	0								

No.	Crop categories	Area under production (Ha 10 ³)	Total Production (MT 10 ³)	Total value US\$ 10 ⁶	
40.	Spinach (Spinacia oleracea)	2.6	50.54	8.65	
41.	Cauliflower (Brassica oleracea)	0.09	0.99	7.13	
42.	Lettuce (Lactuca sativa)	0.2	0.001	0.55	
43.	French beans (Phaseolus sp.)	3.3	46.50	0.02	
44.	Broad bean (Vicia faba var. Major)	0.3	6.09	2.98	
45.	Valor	0.1	0.28	0.11	
46.	Garden peas (Pisum sativum)	11.0	75.40	27.65	
47.	Snow/snap peas (Pisum sativum)	2.6	14.27	7.65	
48.	Bulb onions (Allium cepa)	6.9	88.92	33.24	
49.	Spring onions (Allium sp.)	2.0	26.39	5.66	
50.	Carrots(Daucus carota)	3.2	82.25	20.56	
51.	Radish (Raphanus sativus)	0.01	0.07	0.02	
52.	Okra (Abelmoschus esculentus)	0.6	4.25	1.29	
53.	Karela (Momordica charantia)	0.2	1.03	0.18	
54.	Dudhi/bottle gouard (<i>Lagenaria</i> siceraria)	0.2	2.15	0.44	
55.	Turia (Luffa acutangula	0.1	0.53	0.13	
56.	Ivy gourd/ Tindori (Coccinia grandis)	0.1	0.38	0.10	
57.	Cucumbers (Cucumis sativus)	0.2	3.02	0.92	
58.	Courgette (Cucurbita pepo)/squash	0.8	6.79	2.77	
59.	Pumpkin (Cucurbita maxima)	0.9	18.97	6.27	
60.	Butternut (Cucurbita moschata)	0.5	6.98	2.33	
61.	Tomatoes (Lycopersicon esculentum)	17.2	526.92	207.21	

Appendix 6.1: Cont. Important agricultural crops in Kenya, total national area under cultivation, production and total revenue, 2009

No.	Crop categories	Area under production (Ha 10 ³)	Total Production (MT 10 ³)	Total value US\$ 10 ⁶
62.	Eggplants (Solanum melongena)	0.8	12.77	3.74
63.	Capsicums (Capsicum annum)	1.3	13.36	5.22
64.	Chillies (Capsicum spp.)	1.2	8.70	2.75
65.	Baby corn (Zea mays)	0.6	5.66	2.91
66.	Sweet corn(Zea mays)	0	0.01	0.01
	African Indigenous leafy vegetables			
67.	Mitoo/Slender leaf (<i>Crotalaria brevidens</i>)	0.7	7.01	1.57
68.	Saga/spider plant (<i>Cleome</i> gynandra)	2.5	14.30	2.99
69.	Terere/Amaranth (Amaranthus sp.)	3.7	31.97	7.56
70.	Jute mallow/Corchorus sp.)	1.3	10.86	1.88
71.	Nderema/vine spinach (<i>Bacella alba</i>)	0.1	0.31	0.08
72.	Kahurura/fig leaf gourd (<i>Cucurbita ficifolia</i>)	0.01	0.04	0.01
73.	Managu/broad-leafed African nightshade (Solanum scabrum)	3.1	2.17	5.46
74.	Cow pea leaves (Vigna unguiculata)	15.8	91.01	14.41
	Root crops			
75.	Sweet potatoes (Ipomea batatus)	77.8	1,034.20	316.44
76.	Irish potatoes(Solanum tuberosum)	108.2	2,550.13	370.24
77.	Yams (Dioscorea sp.)	0.9	4.43	*
78.	Arrow roots/Taro (<i>Colocasia</i> esculenta)	2.6	24.90	*
79.	Cassava (Manihot esculenta)	70.4	911.07	*

Appendix 6.1: Cont. Important agricultural crops in Kenya, total national area under cultivation, production and total revenue, 2009

No.	Crop categories	Area under production (Ha 10 ³)	Total Production (MT 10 ³)	Total value US\$ 10 ⁶
	Beverages and stimulants			
80.	Coffee (Coffea arabica)	160.0	47.99	138.96
81.	Tea (Camellia sinensis)	158.4	314.20	*
	Herbs and Spices			
82.	Vanilla (Vanilla planifolia)	0.003	*	
83.	Leeks (Allium ampeloprasum var. porrum)	0.2	1.57	0.3
84.	Coriander (Coriandum sativum)	0.1	0.81	0.26
85.	Celery (Apium graveolens)	0.1	2.88	0.19
86.	Garlic (Allium sativum)	0.6	42.33	3.55
87.	Mint (Mentha sp.)	*	*	16.62
88.	Parsley (Petroselinum crispum)	0.002	0.01	0.01
89.	Turmeric (Curcuma longa)	0.003	0.01	0.01
90.	Ginger (Zingiber officinale)	0.004	0.02	0.01
91.	Rosemary(Rosmarinus officinalis)	0.01	0.24	0.11
	Sugar crops			
92.	Sugar cane (Saccharum officinarum)	154.3	548.21	557.61
	Pesticides			
93.	Pyrethrum (pyrethrin extract) (Chrysanthemum cinerariifolium)	4.1	0.01	1.32
	Fibre crops			
94.	Cotton lint (Gossypium sp.)	*		
95.	Sisal (Agave sisalana)	29.4	19.05	*
	Totals	4,761.07	14,030.21	3,967.65

Appendix 6.1: Cont. Important agricultural crops in Kenya, total national area under cultivation, production and total revenue, 2009

(*) No data recorded.



Appendix 6.2: Kenya political map showing location and the vulnerability ratios to pollinator loss across eight provinces, 2009

Map source: http://www.mapsofworld.com/kenya/.

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To GOD, thank you for the health, the spirit, and the enabling environment. Amen.

CURRICULUM VITAE

CAREER OBJECTIVE

To enhance my career in Agricultural and Biodiversity Research and to acquire sufficient experience that would enable me participate in the environmental and biodiversity conservation, crop production improvements as well as economic development.

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EDUCATION / PROFESSIONAL BACKGROUND

UNIVERSITY EDUCATION

University of Nairobi

- Masters in Agricultural Resource Management (Crop Protection option) 2007
- MSc project: "Assessment of pollinators influence on sunflower yield in Makueni District, Kenya.
- Bachelor of Science in Agriculture (Crop Protection) 2002: Second Class (Upper Division).

SECONDARY EDUCATION

• Aluor Girls' Secondary School (1993 – 1996): KCSE – B (Plain)

SHORT COURSES ATTENDED

- Training in the Safe and Effective Use of Pesticides (2002 April) Agrochemicals Association of Kenya.
- Level One of the IPM (Integrated Pest Management) Practitioners Certificate (31st July-2nd August 2003).
- Bee taxonomy and pollination ecology course at National Museums of Kenya, Nairobi (3rd 28th August 2010).

INFORMATION TECHNOLOGY AND STATISTICS

Conversant and experienced in the following:

- MS Office Packages (Excel, Word, E-mail, and PowerPoint).
- Statistical Packages (SPSS, Genstat).

SEMINARS AND CONFERENCES

- National Museums of Kenya, Nairobi (Nov 11-14, 2004): Conservation and Management of Pollinators for Sustainable Agriculture, Through an Ecosystem Approach, 2nd Kenya Pollinators Initiative Workshop.
- National Museums of Kenya, Nairobi (Feb 14-18, 2005): Termites and Ants Taxonomy Training 2005 Nairobi, Kenya for the Conservation and Sustainable Management of Belowground Biodiversity.
- Kenya Preparatory meeting for the UNCCD Cop 7th to 12th October 2005 at Utalii College, Kenya.
- United Nations Conventions to Combat Desertification (UNCCD) Cop 7 (17th to 28th October 2005) at UN Offices Gigiri Nairobi (Kenya).

SCIENTIFIC PUBLICATIONS

PUBLISHED PAPERS

Kasina JM, Nyamasyo, GN, and Oronje ML (2008) Diversity of sunflower (*Helianthus annus* Nderitu JH, L.) pollinators and their effect on the seed yield in Makueni District, Eastern Kenya. Spanish Journal of Agricultural Research 6 (2): 271-278.

 Nderitu JH, Kasina JM, Nyamasyo GN and Oronje ML (2007) Effects of Insecticide application on sunflower (*Helianthus annus* L.) pollination in Eastern Kenya. World Journal of Agricultural Sciences 3 (6):731-734.

PAPERS IN PRESS AND UNDER REVIEW

- 1. Oronje ML, Hagen M, Gikungu M, Kasina MJ, Kraemer M (2011) Pollinator diversity, behaviour and limitation on yield of Karela (*Momordica charantia* L.: Cucurbitaceae) in Western Kenya. African Journal of Agricultural Research.
- **2.Oronje ML** and Kraemer M (2011) Pollination needs and the role of solitary bees on seed set and quality of slender leaf (*Crotalaria brevidens* Benth: Fabaceae) in Kakamega, Western Kenya. Journal of Pollination Ecology.

REFEREED CONFERENCE PAPERS

 Kasina JM, Nderitu JH, Nyamasyo GN and Oronje ML (2007) Sunflower pollinators in Kenya: Does diversity influence seed yield? 8th African crop science conference, Cairo, Egypt.

PAPERS PRESENTED ON SEMINARS OR TALKS

- Oronje ML and Kraemer M (2011) Pollination needs and the role of solitary bees on seed set and quality of slender leaf (*Crotalaria brevidens* Benth: Fabaceae) in Kakamega, Western Kenya. 10th International Symposium of Pollination, 27th - 30th June 2011, Cholula, Mexico.
- Oronje ML (2011) Effectiveness of pollinators and their potential for crop pollination under greenhouses in Kenya. BIOTA East Africa-KAAD conference, 11th-14th August, 2011, Kakamega, Kenya.
- Kasina MJ, Oronje ML, Kipyab P and Muchira C (2010) Pollination, bee taxonomy and utilization of bioinformatics in pest management. Crop Health Seminar Series, KARI-NARL, 6th October 2010, Nairobi, Kenya.
4. Kasina MJ and **Oronje ML** (2010) Pollination of African traditional leafy vegetables and greenhouse crops. KARI Seminar series, KARI Headquarters, 23rd July, Nairobi, Kenya.

EMPLOYMENT BACKGROUND

• Kenya Plant Health Inspectorate Service (KEPHIS) (November 2006 to May 2008)

Position: Plant Inspector

Responsibilities:

- 1. Inspection of plant materials for phytosanitary compliance for the export and import market.
- 2. Pest identification for effective pest management.
- 3. Pest Risk Analysis (PRA) for market access for agricultural products.
- 4. Reviews and development of inspection protocols for quarantine pests.
- 5. Evaluations on national crop performance trials.
- University of Nairobi at the College of Agriculture and Veterinary Sciences, Plant Science and Crop Protection Department (October 2005 to May 2006)
 - **Position:** Assistant Tutorial Fellow
 - Responsibilities:
 - 1. Preparing teaching syllabus fort the Diploma Crop Protection Class on Bacteriology and Agricultural Entomology.
 - 2. Assisting in the lecturing of the Diploma Crop Protection Class on Bacteriology and Agricultural Entomology.
 - 3. Practical guidance and examination preparation and marking for the Diploma Crop Protection Class on Bacteriology and Agricultural Entomology.

• Dudutech-Homegrown Limited (April 2003–October 2003)

Position: Technical Liaison Officer.

Responsibilities:

1. Undertaking pesticide efficacy trials.

- 2. General pesticides effect on the natural enemies and the pollinators.
- 3. Monitoring the economic damage levels of common pests in the clients' farm and reconciling clients' natural enemies' requirements and the company productions.
- 4. Advising the clients on what amount and when to apply the natural enemies.
- 5. Ensuring that the required favorable conditions exists in the clients farms to maintain the applied natural enemies and reduce the number of spray meanwhile promoting safe pesticide use.

Erklärung (Declaration)

Ich versichere, dass ich diese Arbeit selbständig verfasst habe, keine anderen Quellen und Hilfsmaterialien als die angegebenen benutzt und die Stellen der Arbeit, die anderen Werken dem Wortlaut oder dem Sinn nach entnommen sind, kenntlich gemacht habe.

Diese Arbeit hat in gleicher oder ähnlicher Form keiner anderen Prüfungsbehörde vorgelegen.

Oronje, Mary Lucy Bielefeld, October 2011