EFFECT OF PESTICIDES ON THE TOBACCO SPIDER MITE *Tetranychus evansi* BAKER & PRITCHARD ON TOMATOES IN KENYA.

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DECLARATION

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DEDICATION

This work is dedicated to my parents Thomas and Grace, you taught me faith and endurance, your sacrifice too is immeasurable.

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First I thank God in whom I have my being for making all things beautiful in His time.

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ABSTRACT

The objective of this study was to evaluate the efficacy of the pesticides commonly used by farmers in controlling the Tobacco spider mite *Tetranychus evansi* Baker & Pritchard. Laboratory and greenhouse experiments were carried out to determine the susceptibility of *T. evansi* to dicofol, bifenthrin, propargite, lambda-cyhalothrin, dimethoate and profenofos+cypermethrin using the manufacturer's recommended rates. Ovicidal and adulticidal (contact and residual) assays were done in the laboratory. In the greenhouse, mite leaf damage scores were taken and mite population per cm^2 was determined. Laboratory results showed egg mortalities due to contact effect to be 26.1% in dimethoate treatment and 100% in dicofol, bifenthrin, propargite, profenofos+cypermethrin and lambda-cyhalothrin treatments. The adult mortality due to contact with the pesticides was 100% for dicofol, propargite, bifenthrin and profenofos+cypermethrin treatments whereas mortality for lambda-cyhalothrin was 80%, dimethoate 4% and none in the control. Mortality due to residual effect was 100% for dicofol and was lower for all the other compounds although the mites escaped to the cotton barrier.

Greenhouse studies showed that damage scores and mite populations reduced after the application of dicofol, propargite, bifenthrin, lambda-cyhalothrin and profenofos+cypermethrin whereas for dimethoate and control, the scores continued increasing. There was gradual increase in leaf damage and mite populations per cm^2 in both Dimethoate treated plants and the control over time.

Tobacco spider mites (*T. evansi*) collected from Loitoktok, Kibwezi, Athi-River and Subukia were evaluated for their susceptibility to bifenthrin, profenofos+cypermethrin, dicofol, dimethoate, lambda-cyhalothrin and propargite.

Bifenthrin, profenofos+cypermethrin, dicofol and propargite caused significant mortality in the mites from all the populations tested, however, mortality did not differ significantly between the populations for each of these pesticides ($P<0.05$). Pooled data also showed that mortality did not differ signicantly between dicofol, bifenthrin, propargite and dicofol with average contact adult mortalities of 100%, 99.67%, 99.83% and 99.83% respectively. Lambda-cyhalothrin caused significant mortality in the mites from Kibwezi. However, mites from Loitoktok, Subukia and Athi-River showed significant tolerance. There were significant levels of tolerance to dimethoate in all the populations sampled with the highest adult mortality of 42.5% for the Kibwezi population. Pooled data shows that there was significant difference ($P<0.05$) between lambda-cyhalothrin (65.33%), dimethoate (20.5%), the control (1%) and the rest of the pesticides tested. . Mites from most Kenyan farms respond well to bifenthrin, propargite, profenofos+cypermethrin and dicofol. Dimethoate and lambda-cyhalothrin should be avoided

TABLE OF CONTENTS **Page**

Declaration (ii)

Dedication (iii)

Acknowledgement (iv)

Abstract (v)

Table of contents (vii)

List of tables (ix)

List of figures (ix)

List of appendices (x)

CHAPTER ONE

1 Introduction 1

1.1 Origin and cultivation of tomatoes 1

1.2 Tomato production in Kenya 1

1.3 Spider mite species on tomatoes in Kenya 3

1.4 Problem statement 4

1.5 Justification 5

1.6 Objectives 6

1.7 Hypotheses 6

References 7

CHAPTER TWO

2 Literature review 10

2.1 Tobacco spider mite *Tetranychus evansi* Baker & Pritchard 10

2.2 Biology of *Tetranychus evansi* 10

2.3 Damage and crop loss due to *T. evansi* 11

2.4 Methods of control 12

2.5 Pesticide resistance 14

2.6 Methods used to test for pesticide resistance 16

References 19

CHAPTER THREE

3 Laboratory and greenhouse evaluation of six pesticides for the control

of *Tetranychus evansi* on tomatoes 24

3.1 Abstract 24

3.2 Introduction 25

3.3 Materials and Methods 26

3.4 Results 32

3.5 Discussion 36

3.6 References 38

CHAPTER FOUR

4 Effect of pesticides on geographically isolated field populations of *Tetranychus evansi* in Kenya 40

4.1 Abstract 40

4.2 Introduction 41

4.3 Materials and Methods 41

4.3.1 Collection of mites 41

4.3.2 Identification of mites 42

4.3.3 Culturing of mites 43

4.3.4 Ovicidal tests 44

4.3.5 Adulticidal tests 44

4.4 Results 46

4.5 Discussion 51

References 52

CHAPTER FIVE

5.1 General discussion, conclusions and recommendations 54

5.2 References 56

Appendices 57

LIST OF TABLES Page

Table 1: Tomato production statistics for Kenya (2002-2003 2

Table 2: The contact effect of the pesticides *T. evansi* eggs after six days on leaf discs in the laboratory 33

Table 3: The contact and residual effect of the pesticides on *T. evansi* adult females after 24 hours on treated leaf discs in the laboratory 33

Table 4: Average mite population per cm^2 and damage scores on tomato plants 36

Table 5: Percentage of farmers growing different varieties in different tomato growing regions. 47

Table 6: Percentage of farmers using different pesticides for spider mites control 47

Table 7: Percentage of farms where different spider mites species were collected from 48

Table 8: The contact effect of the pesticides on *T. evansi* egg emergence from different tomato growing regions six days after treatment 49

Table 9: The contact effect of the pesticides on *T. evansi* adult females from different tomato growing regions after 24 hours 49

Table 10: The residual effects of the pesticides from *T. evansi* adult females from different tomato growing regions after 24 hours 50

Table 11: Pooled average effects of the pesticides on the eggs and adult females of *T. evansi* from different tomato growing regions 50

LIST OF FIGURES

Figure 1: The average effect of various pesticides on the tobacco spider mites damage score on tomatoes in greenhouse conditions over time. 35

Figure 2: The effect of various pesticides on tobacco spider mites population on tomatoes in green house conditions over time. 35

LIST OF APPENDICES

Appendix 1: Damage score on a scale of 1-5 (Hussey and Scopes) 57

Appendix 2: A map of the geographical sites where the mites used were sampled from 58

Appendix 3: Pesticide resistance field questionnaire 59

Appendix 4: The GPS co-ordinates of farms where mites were sampled from 60

CHAPTER ONE: INTRODUCTION

1.1 Origin and cultivation of tomatoes.

Tomato, *Lycopersicon esculentum* (Mill), belongs to the family Solanaceae. It originated in tropical Central and South America. It was first domesticated in Mexico and its popularity has increased in the tropics and the subtropics since the end of the nineteenth century (COPR, 1983). Tomato is one of the world's most popular vegetables, with an annual world production of 110 million metric tons in the year 2003 (FAO, 2004). Out of this, 12.6 million metric tons were produced in Africa.

Tomato is grown for its fruit, which contains vitamins A and C. The fruits are eaten raw in salads, cooked as a condiment in stews or made into soup, sauce, juice, ketchup, paste, puree, powder, canned and used unripe in chutneys (COPR, 1983).

1.2 Tomato production in Kenya

Tomato is among the most valuable vegetables in Kenya contributing to the country's gross domestic product KSh. 4.1 billion and 5.1 billion in the year 2002 and 2003, respectively (Table 1.)

Over the years, tomatoes have been produced mainly for domestic consumption. However, some varieties of tomatoes such as the cherry type are now being grown for export market, though their production is low (HCDA, 2002).

Table 1: **Tomato Production Statistics for Kenya (2002/2003)**

Province	Hectarage (Ha)		Production (MT)		Value (Ksh.)	
	2002	**2003**	**2002**	**2003**	**2002**	**2003**
Coast	1572	1098	19048	12303	340,548,000	191,640,000
Eastern	2258	1945	38753	38223	191,640,000	307,823,000
Central	4247	4,314	66422	73,999	448,866,480	1,011,405,000
Nyanza	3931	4,644	76,418	100,869	1,477,600,000	1,854,400,000
Western	1852	1940	30508	30,860	909,277,520	919,172,000
RiftValley	3303	3,949	50312	58,873	578,026,160	645,801,499
Nairobi	77	78	1258	1272	36,942,740	37,482,720
N/Eastern	190	195	2140	2240	128,423,220	134,849,360
Total	**17,430**	**18163**	**284,859**	**318639**	**4,111,324,120**	**5,102,573,579**

Source: Provincial Reports, Ministry of Agriculture Research and Development 2002/2003

Production problems experienced in growth of tomatoes include pests and diseases, high cost of inputs, poor quality seeds and adverse weather conditions. Other problems include uncoordinated and unorganized marketing, exploitation by middlemen and poor production planning leading to over-supply in some months thus very low prices (MoARD, 2001).

The major diseases of tomatoes include late blight (*Phytophthora infestans* Mont. De Bary), early blight (*Alternaria solani* Ell. & Martin), bacterial wilt (*Ralstonia solanacearum* Smith), bacterial canker (*Clavibacter michiganense* subsp. *michiganense* (Smith) Jensen), *Fusarium* wilt (*Fusarium oxysporum* f.sp. *lycopersici* Synder & Hansen) and nematodes (*Meloidogyne* spp.) (Varela *et al.*, 2003). The arthropod pests of importance include the red spider mites (*Tetranychus spp.*), African bollworm (*Helicoverpa armigera* Hübner), whiteflies (*Bemisia tabaci* Gennadius*)* and

aphids (*Aphis* spp). These pests and diseases are also a serious problem in tomato production in other parts of Africa (Varela *et al*., 2003).

1.3 Spider mite species on tomatoes in Kenya

Red spider mites are important pests of tomatoes (MOARD, 2001; Varela, *et al*., 2003.). The most common species of spider mites in Kenya are *Tetranychus evansi* Baker and Pritchard, *Tetranychus urticae* Koch and *Tetranychus cinnabarinus* Boisduval. Until recently, *T. urticae* and *T. cinnabarinus* were the most important spider mites attacking tomatoes in Kenya. However, *T. evansi* identified in the country for the first time in 2001 is more severe than *T. urticae* and *T. cinnabarinus* (Knapp, 2002). It is the most serious dry season pest of tomatoes in the areas where it occurs. Plants are heavily damaged if no control measures are taken (Varela *et al.*, 2003).

According to Kamau (1985), red spider mites, *Tetranychus* spp infest and cause heavy damage to tomato plants with infestation levels of about 25% in Kirinyaga and 10% in Kajiado districts. The percentage of farms infested with *Tetranychus* spp in Nakuru, Kiambu, Kajiado, and Kirinyaga in 1985 was about 25%, 20%, 45% and 65% respectively. Percentage yield losses caused by *Tetranychus* spp in the same districts ranged from 40 to 50%.

A survey done in Kenya in 1998 (Löhr *et al*., 1998) and in 2003 (ICIPE, 2003) reports that *T. urticae,* a species with worldwide distribution and a wider host range, was the predominant species. The report also indicates that red spider mites were only a problem in areas where farmers treated their crops frequently with broad spectrum insecticides. The report also indicates that this situation was expected to change since *T. evansi* was detected in Kenya in May 2001. Kamau (1977), also reported that the

red spider mites (*Tetranychus* spp) is one of the serious pest of tomato wherever it is grown in semi-arid areas under irrigation and in other areas where the crop experiences periods of warm dry spells.

Several pesticides have been used for red spider mite control and the following are commonly used in Kenya: dicofol, lambda-cyhalothrin, dimethoate, cypermethrin, bifenthrin, propargite and sulphur. Other pesticides that are used by a smaller percentage of farmers include tetradifon, endosulfan, omethoate, malathion, alpha cypermethrin, permethrin, fenithrothion, carbosulfan, fenthion, chlorpyriphos, deltamethrin, diazinon and azocyclotin (ICIPE, unpublished survey data).

The chemicals that were used in this study are dicofol (Kelthane), lambda-cyhalothrin (Karate), dimethoate (Danadim), cypermethrin+profenofos (Polytrin) and bifenthrin (Brigade). These chemicals are the ones mostly used by the farmers according to findings from a recent survey which covered Machakos, Mwingi, Kitui, Taita Taveta, Makueni, Kajiado, Kirinyaga, Mbeere, Kwale, Kilifi and Trans-Nzoia districts as recorded below (Table 2) (ICIPE, unpublished survey data).

1.4 Problem statement

The common control strategies of most farmers for vegetable pests, especially mites, rely solely on frequent application of highly toxic acaricides with often inappropriate spraying equipment and dosage. This has resulted in speeding up mite resistance to chemicals faster than most insects due to their short life cycle and high reproductive rates as observed with *T. urticae* (Yoon *et al.*, 2001, James and Price, 2002).

Although many investigations on the toxicity of pesticides to tetranychid mites have been done (Cranham and Helle, 1985), fewer studies on *Tetranychus evansi* have been documented. So far, no effective natural enemy has been found for biological control

of *T. evansi,* hence chemical control still remains the major control strategy. Pesticide resistance has been documented in *T. urticae* (Cranham and Helle, 1985, Dennehy and Granett, 1984, Goodwin *et al.*, 1995, Jensen and Mingochi, 1988, Kolmes *et al.*, 1994,), which is the other important species in tomato production in East Africa (Varela *et al.*, 2003). *T. evansi* is gaining importance in tomato production (Varela *et al.*, 2003) and as Smith (1970) suggested, it is an essential part of pest control procedures to monitor continuously the level of pesticide resistance in major pests. However, nothing has been documented on pesticide resistance in *T. evansi.*

1.5 Justification

The red spider mite *T. evansi* is a serious pest of tomatoes. It causes more damage than the two-spotted spider mite *T. urticae* (Knapp *et al.*, 2003). Due to the absence of an effective natural enemy in Africa and because few new acaricides are under development, and even fewer are acceptable in developing integrated pest management programs, it is important to both preserve and maximize the effectiveness of available products (Marshall and Pree, 1991).

The most commonly used acaricides in Kenya are dicofol, lambda-cyhalothrin, dimethoate, propargite, bifenthrin and cypermethrin. However, the susceptibility of *T. evansi* to these compounds is not known. The resistance status of *T. urticae* and some other important mite species to acaricides is well documented (Cranham and Helle, 1985; Whalon & Mota-Sanchez, 2000). Given the increasing pest status of *T. evansi* in Kenya and in Africa, there is need to develop control packages that are sustainable. This will include use of selective pesticides that will not affect non-target organisms and are effective. It is therefore expedient that studies to determine the susceptibility of *T. evansi* to the commonly used acaricides and insecticides are carried out in order

to gain information on the possible resistance by *T. evansi* to these pesticides, and as a result, only those pesticides that show continued toxicity are recommended for use by farmers. It is also important to determine if resistance is a problem in a specific location or time in order to improve pesticide choice (Dennehy and Granett, 1984). This will in turn reduce the possibility of pest resurgence associated with high use of pesticides. By eliminating unnecessary pesticide application, production costs will be reduced.

1.6 Objectives

1.6.1 Overall Objective

To evaluate the efficacy of the pesticides commonly used by farmers in controlling the tobacco spider mite *Tetranychus evansi* Baker & Pritchard.

1.6.2 Specific objectives

i. To determine the effect of bifenthrin, dicofol, propargite, lambda-cyhalothrin, dimethoate and profenofos+cypermethrin on laboratory maintained population of *Tetranychus evansi* in laboratory and greenhouse conditions

ii. To determine the effect of bifenthrin, dicofol, propargite, lambda-cyhalothrin, dimethoate and profenofos+cypermethrin on geographically isolated field populations of *Tetranychus evansi*.

1.7 Hypotheses

(i) Specific acaricides show higher toxicity to *Tetranychus evansi* compared to broad-spectrum insecticides.

(ii) Continuous exposure of T. evansi to the same pesticide over time leads to resistance

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CHAPTER TWO: LITERATURE REVIEW

2.1 Tobacco spider mite, *Tetranychus evansi* Baker & Pritchard

Tetranychus evansi Baker & Pritchard was first recorded as *Tetranychus marianae* McGregor from north-eastern Brazil and Mauritius (Moutia, 1958) and later redescribed as *T. evansi* (Baker and Pritchard, 1960) from material collected in Mauritius. The mite was found in *Solanum nigrum* L, *Lycopersicon esculentum* Mill, *Solanum melongena* L, *Artemisia douglasiana* Bess, *Nicotiana glauca* Graham, *Phacelia* sp., *Rosa* sp., and lily of the valley vine, *Salpichora rhomboidea* Miers (Harper 1966, Oatman et al.1967a). It also attacks potatoes, peppers, wild gooseberry and the bitter apple (Varela *et al*. 2003).

T. evansi was first reported in continental Africa from tobacco in Zimbabwe in 1979 (Blair, 1983) and now occurs in Zambia, Malawi, Namibia and Mozambique (Knapp *et al*, 2003), South Africa (Meyer, 1996), Congo (Bonato, 1999), Morocco and Tunisia (Bolland *et al.* 1998). It was first reported in Kenya in May 2001 in a laboratory culture at ICIPE which was started from mites collected at Mwea irrigation Scheme in Central Kenya (Knapp *et al*., 2003).

2.2 Biology of *Tetranychus evansi*

Tetranychus evansi has a development time of 8 days from egg to adult at a temperature range of 24-26^{0}C. Field collected females lay an average of 16 eggs per day during a five day period. The preferred hosts were killed within 3-5 weeks of initial infestation (Oatman *et al.,* 1967a and b). The short life cycle and high number of eggs laid give *T. evansi* the power to increase rapidly to very high numbers on

suitable hosts in a favourable environment (Qureshi *et al*., 1969). Females live for about 26-34 days whereas males live an average of 25 days. Wind is probably the most important agent in the local distribution of *T. evansi*. At high populations, the adults spin copious webs and form clusters on the tips of heavily infested, defoliated host plants from which they are probably easily dispersed by air currents (Qureshi *et al*., 1969).

Eggs are 0.1mm in diameter, whitish and are laid singly on the underside of the leaf. They hatch after 4 to 7 days into larvae which are six-legged, pinkish in colour and slightly larger than the egg. This lasts for 3 to 5 days then develops into the nymphal stage which lasts 6 to 10 days. There are two nymphal stages, they have four pairs of legs and are reddish. Adult females are oval, about 0.3-0.5mm long, orange-red with reddish legs and an indistinct dark blotch on each side of the body. Adult males are light orange and smaller than the females. Adults spin fine strands of silk that form an open web above the leaf surface. The adult female may lay over 100 eggs. All active stages feed together on the lower sides of the leaves (Keizer and Zuurbier, 2000; Meyer, 1996).

2.3 Damage and crop loss due to *T. evansi*

Damage first appears as stipples that later give way to yellowish and silvery appearance. The mites prefer the lower surface of leaves but in severe infestations occur on both leaf surfaces as well as on fruits and stems. As the population increases, the mites may completely cover the plants with webbing. Plants with a high infestation can be covered with an orange cloud of mites. Eventually the leaves turn brown and die. These mites can kill plants very rapidly under hot and dry conditions

(Meyer, 1996, Varela *et al*., 2003). High infestations that occur later in the crop season lead to production of smaller and lighter fruits. It may also cause speckling of the fruits (Varela *et al*., 2003).

2.4 Methods of Controlling the Tobacco spider mite

Farmers are known to use a number of methods in controlling pests. Cultural control, the modification of management practices that make the environment less favourable to pest reproduction, dispersal and/or survival are commonly used. There are several methods that reduce mite population and they are used in integrated control. Removal and burning of infested plants during the early stages of infestation of mites when they concentrate on a few plants has been found successful. The separation of infested crops and newly planted crops or nursery areas and the burning or removal of infested crop residues and weeds also help to minimize the problem (Keizer and Zuurbier, 2000). Mites prefer dry and hot conditions. Influencing the microclimate by reducing the spacing and applying overhead irrigation has been found to suppress the mite populations. However, this could also enhance fungal diseases so care should be taken. Water and nutrient stress should be avoided since this has been proven to increase mite populations. Applying mulch and incorporating organic matter into the soil can improve the water holding capacity and reduce evaporation, thus avoid water stress (Keizer and Zuurbier, 2000).

Biological control, by means of natural enemies is cheap, effective, permanent, and non-disruptive of the ecosystem. Moraes & McMutry (1985, 1986) investigated the suitability of *T. evansi* as a prey for *Phytoseiulus persimilis* Athias-Henriot and seven other phytoseiid mites. None of these was an effective predator of *T. evansi* as their

oviposition and survivorship were very low on this prey. To date, only small staphylinid beetles (*Oligota* spp) are known to feed on this pest (Varela *et al*, 2003). However, rearing this predator is difficult due to its delicate larvae and pupation that occurs in the soil (Knapp, pers comm.). Pathogenic fungi have been used for biological control of *T. evansi* with considerable levels of success using *Beauveria bassiana* and *Metarhizium anisopliae* (Wekesa, 2004). Botanicals have also been used and work by Kithusi (2005) reports some commercial neem formulations to have high potential for the control of T. evansi.

Varietal resistance is a major element in integrated control of spider mites; it is the least expensive and the easiest technique for plant protection. It has been tried in beans, cucumber, tomato and strawberry (Van Impe and Hance, 1993). In Brazil, some promising genotypes resistant to *T. evansi* have been identified (Varela *et al*., 2003) and studies are underway to determine the use of varietal resistance in *T. evansi* control.

Chemical pesticides are the most frequently used and the most effective pest control tools. Spider mites rapidly develop resistance to pesticides, particularly when they are used for several consecutive seasons. Rotation of acaricides with different chemical compositions is necessary to avoid or delay development of resistance. Preventive application and application of dosages lower than recommended should be avoided since this may lead to resistance. Use of specific acaricides at appropriate doses and times of application are essential and use of broad spectrum insecticides should be avoided as much as possible (Varela *et al*., 2003).

Bifenthrin (Brigade) and lambda-cyhalothrin (Karate) are insecticides with acaricidal properties. They are pyrethroids from the trifluoromethyl group. Bifenthrin is used in formulations such as emulsifiable concentrate, soluble concentrate and as a wettable powder. It has contact and stomach action. It acts as an axonic poison by interfering with the sodium channels at peripheral and central nervous systems stimulating repetitive nervous discharges leading to paralysis. Karate is an emulsifiable concentrate with non-systemic contact and stomach action and repellent properties. It acts as an axonic poison by interfering with the sodium channels at peripheral and central nervous systems stimulating repetitive nervous discharges leading to paralysis. It has rapid knock down and long residual activity (Kidd and James, 1991). cypermethrin+profenofos (Polytrin) is a pyrethroid with insecticidal and acaricidal action (Syngenta, 2005). It is non-systemic with contact and stomach action, and anti-feeding action. It has good residual activity. Dimethoate (Dimethoate; Danadim) is an organophosphate with insecticidal and acaricidal properties. It is a cholinesterase inhibitor with systemic, contact and stomach action. It interrupts the transmission of nerve impulses. Dicofol (Kelthane) is an organochlorine. It is an acaricide with contact action and it kills eggs and all active stages of mites. Propargite (Omite) is a sulfite ester (Morse *et al.*, 1987) and is an acaricide. It has contact, non-systemic action with long residual activity (Kidd and James, 1991.)

2.5 Resistance of spider mites to pesticides

Strict chemical control of spider mites is difficult because of their propensity to develop resistant strains (Croft and van de Baan, 1988; Reissing *et al.*, 1986). Pesticide resistance evolve via metabolic detoxifying mechanisms (Dennehy and Granett, 1984; Dennehy *et al.*, 1987) to behavioral responses such as dispersal

behaviors (Franklin and Knowles, 1984), changes in feeding activity (Kolmes *et al*., 1990) and influences upon oviposition site selection (McPherson *et al*., 1989).According to the database of arthropods resistance to pesticides (Whalon and Mota-Sanchez, 2000), *T. urticae* is reported to be resistant to 275 pesticides including bifenthrin, dicofol, dimethoate, profenofos and propargite. *T. urticae* resistance to dicofol was also reported by Dennehy and Granett (1984); Karban and Zalom (1998); Kolmes *et al.* (1994) and Rossi and Conti (1997), resistance to bifenthrin by Kolmes *et al.* (1994) to propargite by Goodwin *et al*. (1995) and to dimethoate by Jensen and Mingochi, (1988). Mansour and Plaut (1979) reported resistance to dicofol in the carmine spider mite *Tetranychus cinnabarinus* (Boisduval) collected from field and ornamental crops and found resistance a factor of 167 fold. Studies done by Bynum *et al.* (1997) suggest that metabolic degradation and target site insensitivity may be involved in mite resistance. General esterases and glutathione S-transferases (GST) are related to and possibly responsible for changes in susceptibility of the twospotted spider mite to several insecticides particularly the synthetic pyrethroids (Yang *et al*, 2001). Other studies showed that *T. urticae* are much more resistant to residual applications of dicofol than to topical applications (Smith, 1960) and similar observations with parathion were documented by Walker *et al*. (1973). Not much has been documented on pesticide resistance in *T. evansi* hence the wide reference to *T. urticae* that is a closely related species (Knapp *et al*, 2003) and a widely documented species in Eastern Africa (Meyer, 1996). Blair (1989) tested 62 acaricide formulations against *T. evansi* on tobacco in the laboratory. He found that control with dimethoate and other thiophosphates was poor and these observations were confirmed by field results.

2.6 Methods used to test for pesticide resistance in spider mites.

2.6.1 Slide-dip method.

In the slide-dip method, female mites are affixed by their dorsal surfaces on double-sided adhesive tape attached to a microscope slide (Dennehy and Granett, 1982; Dittrich, 1962; Kabir *et al*., 1993). Slides are dipped for 5 seconds in the pesticide solutions. After dipping, the slides are left to dry, excess fluid carefully blotted off the slides using a filter paper (Dittrich, 1962; Kabir *et al*., 1993). The treated slides are placed in covered plastic trays lined with slightly moistened paper towels and transferred to an incubator. After 24 hours, survivors are counted (Dittrich, 1962). The mortality criterion is the inability to move a leg when lightly prodded with a fine brush (Kabir *et al*., 1993).

2.6.2 Cage spray method

This method was outlined by Dittrich (1962). Mites are transferred into fine mesh cages glued to the surface of primary plant leaves. Mites, chosen under the microscope for uniformity in size and appearance, are transferred by a pneumatic collector to the cages. The number of test individuals in each cage is ascertained under the microscope then sprayed using a constant amount of liquid, determined by the time and pressure used for spraying.

2.6.3 Leaf disc direct – Potter tower method (LDD-PT)

Leaf discs are placed upside down in plastic Petri dishes containing moistened cotton wool or a semi-solid agar pad. Adult females are transferred on to each leaf disc using a fine brush. After release onto the leaf discs, the mites are left undisturbed for at least an hour at room temperature. Individual Petri dishes are then sprayed with 2ml of pesticide solutions in a Potter tower operated at 55kPa and allowing 10 seconds settling period. This results in a wet deposit of 1.25±0.01 $mgcm^{-2}$ (Kabir *et al*., 1993).

After 2 hours, the number of dead mites is taken and mortality is determined by getting the percentage of dead mites compared to the total number of treated mites. Resistance is determined by the proportion of mites that survive 24 hours post treatment.

2.6.4 Leaf disc residue – Potter tower method (LDR-PT).

Leaf discs are placed upside down in plastic Petri dishes containing moistened cotton wool or a semi-solid agar pad. Individual Petri dishes are then sprayed with 2ml of pesticide solutions in a Potter tower operated at 55kPa and allowing 10 seconds settling period. The spray deposit is left to dry at room temperature for 15 minutes. Adult females are transferred on to each leaf disc using a fine brush (Kabir *et al.*, 1993).

2.6.5 Leaf disc residue – dipping method (LDR-D)

Leaf discs are dipped for 5 seconds in pesticide solutions then placed upside down in plastic Petri dishes containing moistened cotton wool or a semi-solid agar pad. The Petri dishes are left to dry at room temperature for 15 minutes. Adult females are transferred on to each leaf disc using a fine brush (Kabir *et al.,* 1993).

2.6.6 Petri dish residue – Potter tower method.

Clear plastic Petri dishes with fitting lids measuring 50mm diameter and 9 mm deep are used. The internal surfaces of lids and bases of each Petri dish are sprayed with 2ml of the pesticide solutions under the Potter tower. Following treatment, the Petri dishes are left uncovered for half an hour to dry at room temperature. A small plug of 1.5 % w/v agar is placed on the internal surface of each lid to maintain sufficient humidity inside the dishes. Adult mites are transferred to each dish. The prepared dishes are placed in covered plastic trays in an incubator

2.6.7 Petri dish residue – rinsing method (PDR-R)

This method was described by Kabir *et al.* (1993) and Dennehy *et al*. (1987) referred to it as a practitioner- assessable (PA) bioassay. Clear plastic Petri dishes with fitting lids measuring 50mm diameter and 9 mm deep are used. 2ml of the pesticide solution is placed in each dish, the lid closed tightly, and the dish gently swirled for 5 seconds in both an upright and inverted position. Excess solution is poured off and the dish allowed to dry at room temperature for 30 minutes. Adult mites are then transferred to each dish using a fine brush.

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CHAPTER THREE

LABORATORY AND GREENHOUSE EVALUATION OF PESTICIDES FOR THE CONTROL OF *Tetranychus evansi* ON TOMATOES

3.1 Abstract

Laboratory and greenhouse experiments were carried out to determine the susceptibility of *T. evansi* to dicofol, bifenthrin, propargite, lambda-cyhalothrin, dimethoate and cypermethrin+profenofos using the manufacturer's recommended rates. Ovicidal and adulticidal (contact and residual) assays were done in the laboratory. In the greenhouse, mite leaf damage scores were taken and mite population per cm^2 was determined. Laboratory results showed that egg mortality due to contact effect was 0% in the control, 26.1% in dimethoate treatment and 100% in dicofol, bifenthrin, propargite, profenofos+cypermethrin and lambda-cyhalothrin treatments. Adult mortality due to contact with the pesticides was 100% for dicofol, propargite, bifenthrin and profenofos+cypermethrin treatments whereas mortality for lambda-cyhalothrin was 80%, dimethoate 4% and 0% for control. Mortality due to residual effect was 100% for dicofol and lower for all the other compounds although the mites drowned in the cotton wool.

Greenhouse results showed that damage scores and mite populations per cm^2 reduced after application of dicofol, propargite, bifenthrin, lambda-cyhalothrin and cypermethrin+profenofos. There was gradual increase in leaf damage and mite populations per cm^2 in both dimethoate treated plants and the control. All the pesticides tested except dimethoate are effective in control of *T. evansi* using the manufacturer's recommended rates.

3.2 Introduction

Chemical control is the main control strategy used to manage red spider mites in Kenya. The synthetic pesticides being used against *T. evansi* in Kenya include dicofol, lambda-cyhalothrin, dimethoate, profenofos+cypermethrin, bifenthrin, propargite and sulphur (ICIPE, unpublished). There are a few other acaricides that are being developed for *T. evansi* control with success like spiromesifen, which is mainly effective as an ovicide (Machini, 2005).

Frequent use of acaricides often result in speeding up mite resistance to chemicals faster than in most insects due to their short life cycle and high reproductive rates (Cranham and Helle, 1985). Although many studies on the toxicity of pesticides to tetranychid mites have been done, fewer studies on *T. evansi* have been documented. Among the acaricides found to be promising in *T. evansi* control are amitraz, binapacryl, monocrotophos, profenofos, triazophos and dicofol (Blair, 1989).

Given the increasing pest status of *T. evansi* in Kenya (Varela *et al*. 2003), there is need to determine a suitable chemical control package. It is therefore expedient that studies to determine the susceptibility of *T. evansi* to the commonly used acaricides and insecticides are carried out in order to gain information on the toxicity of the chemicals and as a result, only those pesticides that show continued toxicity are recommended for use by farmers. This will reduce the possibility of pest resurgence associated with high use of pesticides and by eliminating unnecessary insecticide application, production costs will be reduced.

This study was carried out to determine the susceptibility of laboratory reared *T. evansi* to dicofol, bifenthrin, lambda-cyhalothrin, propargite, dimethoate and

profenofos+cypermethrin in the laboratory and in greenhouse conditions. The commonly used pesticides by the farmers according to previous survey findings were chosen.

3.3 Materials and methods

3.3.1 Laboratory evaluation of pesticides for the control of *T. evansi* on tomato leaves

3.3.1.1 Growing of tomatoes for the laboratory experiments

All the laboratory experiments were carried out at ICIPE Red Spider Mite laboratories. Tomato plants used to rear the mites and for leaf discs were grown in the greenhouse at ICIPE. The tomato variety used for the laboratory experiments was Cal-J. This tomato variety was chosen because it is susceptible to *T. evansi* infestation and is also commonly grown in Kenya for fresh market.

Tomatoes were raised in germination trays and transplanted in pots (14 x 15 x 8 cm) in a green house at ICIPE premises. The planting media used was three parts of red soil, two parts of manure and one part of sand. The plants received the required management practices that are watering; weeding and top dressing with calcium ammonium nitrate (CAN) at a rate of 3 grams per pot. Upon reaching the required size (one month after transplanting) leaves were harvested to make the leaf discs for laboratory experiments.

3.3.1.2 Spider mite culture

A pure culture of *T. evansi* that had been collected from Mwea in 2001 and maintained in the laboratory at ICIPE without exposure to any pesticides was used in

this study. The mites were reared in the laboratory at room temperature (25.0 ± 2^0C), relative humidity 70 ±10% under L12:D12 photoperiod on tomato plants 4 - 6 weeks after transplanting. To maintain the mite stock, host plants (tomatoes, Money Maker variety) were sown regularly and twice a week 3 to 4 weeks old were placed next to aging plants to enable mites to move to the fresh plants. Individual mites used for the bioassay tests were collected and transferred from infested leaves to leaf discs using a fine hairbrush. Care was taken not to injure the mites, as they are soft bodied.

3.3.1.3 Ovicidal tests

Commercial samples of Polytrin (profenofos Q 400g/l + cypermethrin 40) 440 EC; Brigade (bifenthrin 25g/l) 025EC; Karate (lambda-cyhalothrin 17.5g/l) 1.75EC; Dimethoate (dimethoate 400g/l) 40% EC; Omite (propargite 57%) 57EC and Kelthane EC (dicofol 18.5%) were used in the experiments.

For each pesticide, 20 *T. evansi* females from the laboratory colony were transferred to tomato leaf discs (25mm) placed on moistened cotton in petri dishes. The females were allowed to oviposit for 24 hours after which the mites were removed and the number of eggs adjusted to twenty. Pesticide solutions were prepared using the manufacturer's recommended rates for each pesticide as follows:

Profenofos+cypermethrin – 1.5ml per litre; bifenthrin – 2.5ml per litre; lambda-cyhalothrin - 2.5ml per litre; dimethoate - 1ml per litre; propargite – 1ml per litre; dicofol – 2.5ml per litre.

A sticker (Aquawet) was added onto the solutions at a rate of 0.5ml per litre to improve their sticking ability. The pesticides mentioned above were chosen for this study in order of the list of commonly used pesticides for red spider mites control in

Kenya as recorded from a survey carried out in 2000 (ICIPE, unpublished).

The leaf discs containing the eggs were dipped in the pesticide solutions and water plus sticker as a control for five seconds (Keena et al., 1991). The treatments were replicated six times. The treated leaf discs were placed upside down in plastic petri dishes containing moistened cotton wool. They were then left on the bench to dry for half an hour before being put into plastic boxes. Saturated solutions of potassium carbonate were used to maintain humidity conditions (Winston and Bates, 1960) in the plastic boxes (22 by 33 by 7cm) in which the trays containing the petri dishes were placed. The boxes were covered and kept in the incubator at 25^0C under continuous light until toxicity was assessed. The experiment was arranged in a complete randomized design (CRD). The leaves were examined after 4 days for hatched larvae and were further examined daily for another four days. Ovicidal mortality was determined by comparing the number of unhatched eggs with the original number of post treatment eggs (Agnello, *et al.,* 1994).

Adulticidal tests.

Two bioassay methods modified leaf direct method (LDD) and leaf disc residue – dipping (LDR-D) were used in this study. The two methods are a modification of those described by Kabir *et al*., (1993)

In the modified leaf disc direct method, chemical solutions were prepared using the manufacturer's recommended rates as described in ovicidal tests above. Twenty *T. evansi* adult females of the same age from the laboratory culture were transferred onto the lower side of 25mm diameter tomato leaf discs using a fine brush. The leaf discs containing the mites were dipped in the pesticide solutions for 5 seconds and the discs

placed upside down in plastic petri dishes containing moistened cotton wool. This was done for profenofos+cypermethrin – 1.5ml per litre; bifenthrin – 2.5ml per litre; lambda-cyhalothrin - 2.5ml per litre; dimethoate - 1ml per litre; propargite – 1ml per litre; dicofol – 2.5ml per litre and water plus sticker as control with six replications each. The petri dishes were left uncovered for half an hour to allow the leaves to dry. Saturated solutions of potassium carbonate were used to maintain humidity conditions in plastic boxes (22 by 33 by 7cm) in which the trays containing the petri dishes were placed. The boxes were covered and kept in the incubator at 25^0C. The treatments were arranged in a completely randomized design (CRD). The mites were observed after 24 hours and recorded as dead or escaped for those that were trapped in the cotton wool.

For the leaf disc residue – dipping method, the procedure in LDD method was repeated with the exception that the leaf discs were dipped first in the pesticide concentrations for 5 seconds. They were placed in petri dishes and allowed to dry at room temperature. The mites were then transferred onto the discs. The mites were observed after 24 hours and recorded as dead or escaped for those that were trapped in the cotton wool.

For both methods, mortality was calculated by getting the number of mites dead divided by the number of mites treated excluding the mites which escaped and got trapped in the cotton wool. The data was statistically analysed using ANOVA and means separated using Student-Newman-Keuls (SNK) test (SAS, 1990). Percent mortality data was arc-sin transformed before analysis. Correction for control

mortality was done where mortalities in the control were appreciable using Abbott's formula (1925):

Pt = Po-Pc/ 100-Pc x 100, where

Pt = corrected mortality,

Po = observed mortality and

Pc = control mortality. (All in percentages).

3.3.2 Green house evaluation of pesticides for the control of *Tetranychus evansi* on tomato leaves

Tomato variety Cal-J seeds were sown in germination trays in a greenhouse at International Centre of Insect Physiology and Ecology (ICIPE) premises, Kasarani, Nairobi, Kenya.

Three weeks after germination, 84 seedlings were transplanted in plastic pots (25x32x20 cm) filled with a mixture of topsoil, sand and manure at a ratio of 2:1:1 v/v and double ammonium phosphate (DAP) at a rate of 3g per pot. The potted plants were then taken to a greenhouse at Jomo Kenyatta University of Agriculture and Technology (JKUAT) (01º 01'S 37º 06'S), at 1600m (asl) (Jaetzold & Schmidt, 1983). The potted plants were then placed on benches and the experiment was arranged in a complete randomized design where four potted tomatoes represented one plot. Seven treatments were randomly assigned to each plot replicated three times.

The treatments and the concentrations used were as in 3.2.1.3. The plants were watered as required and calcium ammonium nitrate (CAN) at a rate of 3 grams per pot was applied one month after transplanting. Pruning was done weekly by pinching off laterals leaving one main stem to grow. Staking was done using one-metre long poles

and the tomato stems were loosely tied on the posts using a sisal twine. The greenhouse temperatures ranged from 17-35^{o}C and mean relative humidity was 52%.

Three weeks after transplanting, each plant was artificially infested with 100 adult *T. evansi* mites of both sexes by directly picking using a fine hairbrush from a colony maintained on a tomato plants reared in the laboratory at ICIPE. The mites were evenly distributed on all the leaves and were allowed 21 days to establish and multiply before treatments were administered. Two sprayings were done at 3 weeks and 5 weeks after infestation and the tomatoes plants were sprayed outside the greenhouse using a hand sprayer (1.5 litres) to avoid spillage between the treatments.

Leaf damage index (LDI) and the number of mites per leaf area (cm^2) were determined. Leaf damage index (LDI) was established visually using modification of a scale described by Hussey and Scopes (1985). The visual rating scale used for leaf damage was 1 to 6 where 1 = no damage, 2= 1-15%, 3= 20-30%, 4= 35-50%, 5= 55-70%, 6= 80-100% of leaf damaged (Appendix 3). The initial leaf damage score was done just before the first treatment (3 weeks after infestation) and there after was done every week. The score was done on ten plants per treatment and one leaf per section from top, middle and lower sections of each was assessed and the mean leaf damage was calculated.

All the motile stages of the red spider mites were counted every two weeks with the first count being done one week after treatment application. Three plants were sampled per treatment and from each plant, three terminal leaflets were obtained from the top, middle and lower sections of the plants. These were kept separately in the

labeled paper bags and carried in a cool box maintained at 4ºC. Mite counting was done at ICIPE laboratory using a microscope and a tally counter. After counting, the sampled leaves were returned in the paper bags and taken back to a laboratory at JKUAT for determination of leaf area (cm^2) using leaf area meter.

Calculations were made to establish the average number of mites per cm^2 of the leaf area. The data was analysed using ANOVA, and the counts were log-transformed before analysis. The populations were statistically compared using Student-Newman-Keuls (SNK) test.

3.4 Results

3.4.1 Susceptibility of *T. evansi* to dicofol, bifenthrin, propargite, lambda-cyhalothrin, dimethoate and profenofos+cypermethrin in laboratory conditions

There were no egg emergence in the profenofos+cypermethrin, dicofol, propargite, lambda-cyhalothrin and bifenthrin treated leaves and mortality due to these treatments was 100% (Table 2). Eggs treated with dimethoate showed tolerance with egg mortality being 26% (Table 2).

Adult contact mortality was highest in the dicofol, profenofos+cypermethrin, bifenthrin and propargite treatments. Dimethoate and Control recorded the lowest mortality of 3.3% and 0% respectively.

Mortality due to residual effect was generally lower than the mortality due to contact effect of the pesticides except with dicofol and lambda-cyhalothrin treatments where residual and contact mortality remained constant at 100% and 80.8% respectively. Mortality due to the residual effect of Dimethoate and the control were both 0% (Table 3).

Table 2: The contact effect of the pesticides on *T. evansi* eggs after six days on the leaf discs in the laboratory.

Pesticide	Egg mortality(%) ± SE
Bifenthrin (2.5ml/l)	100.0±0.0a
Profenofos+cypermethrin (1.5ml/l)	100.0±0.0a
Dicofol (2.5ml/l)	100.0±0.0a
Propargite (1ml/l)	100.0±0.0a
Lambda-cyhalothrin (2.5ml/l)	100.0±0.0a
Dimethoate(1ml/l)	26.1±1.1b
Control (water)	0.0±0.0c

Means followed by the same letter within one column are not significantly different (SNK-test, P<0.05).

Table 3: The contact and residual effect of the pesticides on *T. evansi* adult females after 24hr on treated leaf discs in the laboratory.

Pesticide	Adult mortality (%) ±SE	
	Contact	Residual
Dicofol	100.0±0.0aA	100.0±0.0aA
Propargite	99.2±0.8aA	84.2±3.0bB
Profenofos+cypermethrin	100.0±0.0aA	56.7±12.7cB
Bifenthrin	100.0±0.0aA	36.7±8.2cB
Lambda-cyhalothrin	80.8±4.0bA	80.8±4.4bA
Dimethoate	3.3±1.7cA	0.0±0.0dA
Control (water)	0.0±0.0cA	0.0±0.0dA

Means followed by the same lower case letter within one column are not significantly different (SNK-test, P<0.05). Means followed by the same upper case letter within one row are not significantly different.

3.4.2 Susceptibility of *T. evansi* to different pesticides under greenhouse conditions.

Leaf damage scores showed a small increase in all the treatments one week after their application. Damage scores started declining one week later for dicofol, propargite, bifenthrin, lambda-cyhalothrin and cypermethrin+profenofos. By the fourth scoring, the score for all the treatments except for the control and dimethoate was one and below (Fig. 1). There was gradual increase in leaf damage in the dimethoate treated plants and the control plots over time (Table 4).

Populations increased up to 25.4 mites/cm^2 in the control by the sixth week up from 3.3 mites/cm^2 and in the dimethoate treated plants they increased to 13.9 mites/cm^2 up from 1.6 mites/cm^2 compared to profenofos+cypermethrin, lambda-cyhalothrin, propargite, dicofol, bifenthrin where the mites were 1.1, 0.1, 0.5, 0.2 and 0.04 mites/cm^2 respectively (Fig. 2, Table 4).

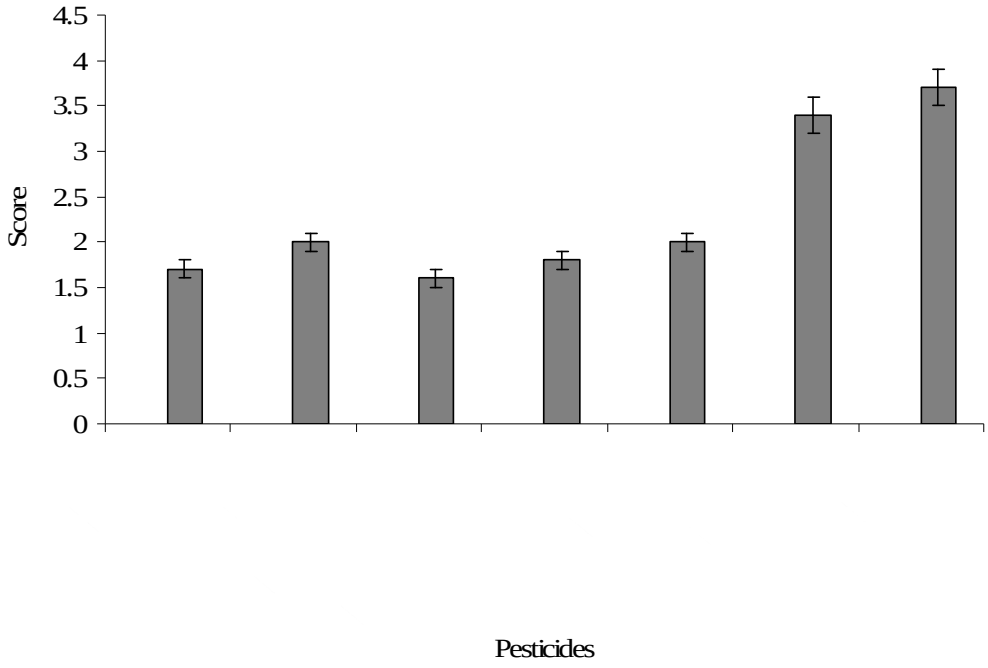

Figure 1: The average effect of various pesticides on the tobacco spider mites (*T. evansi*) damage score on tomatoes in green house conditions.

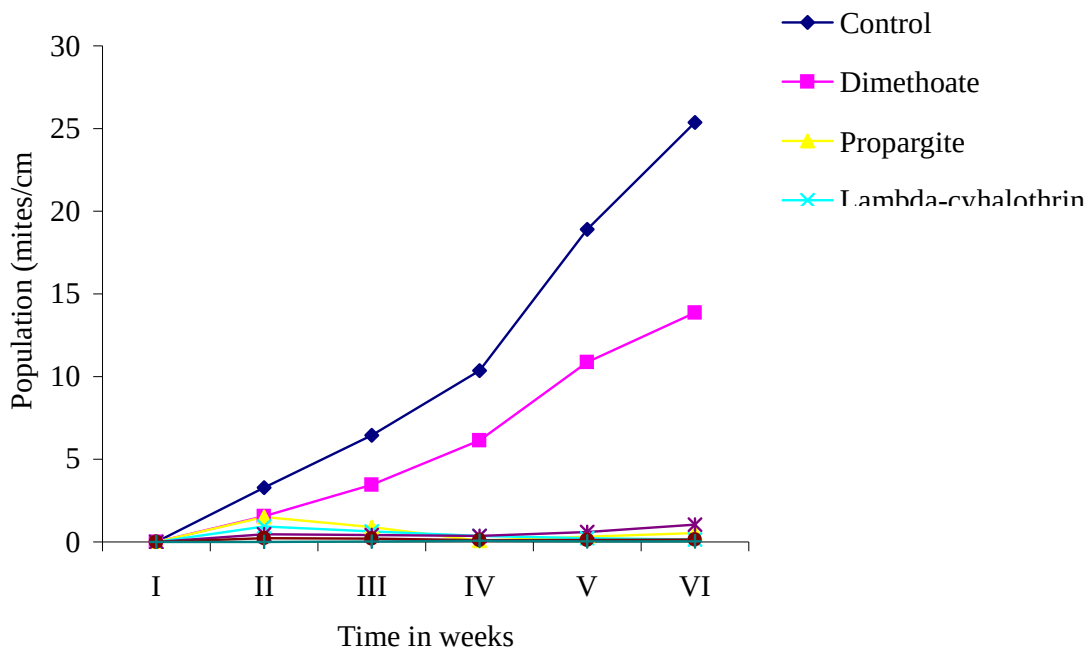

Figure 2: The effect of various pesticides on the tobacco spider mites (*T. evansi*) populations on tomatoes in green house conditions over time.

Table 4: Average mite population per cm^2 and damage scores n tomato plants infested with tobacco spider mites (*T. evansi*) and treated with various pesticides under greenhouse conditions.

Pesticide	Mite population ± SE	Damage score ± SE
Control	12.0 ± 2.3a	3.7 ± 0.2a
Dimethoate	8.1 ± 2.1b	3.4 ± 0.2b
Propargite	0.7 ± 0.2c	2.0 ± 0.1c
Profenofos+cypermethrin	0.7 ± 0.3c	1.8 ± 0.1c
Lambda-cyhalothrin	0.5 ± 0.3c	1.6 ± 0.1c
Dicofol	0.2 ± 0.1c	2.0 ± 0.1c
Bifenthrin	0.0 ± 0.0c	1.7 ± 0.1c
F-value	28.7	52.2

Within column means followed by the same letter are not significantly different (SNK-test, $P<0.05$)

3.5 Discussion

Dicofol, bifenthrin, propargite, profenofos+cypermethrin and lambda-cyhalothrin were effective in the control of the tobacco spider mites both as contact ovicides and adulticides. They also had significant residual effects on the adult mites. This agrees with the manufacturer's labels. Bifenthrin, propargite and profenofos+cypermethrin could be having strong repellence properties due to a high number of mites escaping from the treated surface and drowning in the moist cotton wool when they were exposed to residues of these pesticides as Koehler and Tucker (2003) listed bifenthrin and cypermethrin to be among the pesticides that can be used as repellents against termites. Dimethoate however showed very little effect on the mites both as an ovicide and as an adulticide and this confirms farmer complaints in the fields and studies done by Blair (1989), which showed that most thiophosphates had no significant effects on *T. evansi* in the laboratory. Jensen and Mingochi (1988) reported problems of control of *T. evansi* using dimethoate and other organophosphates but found cyhexatin and propargite to effectively control the mites. This clearly agrees with the findings of the present study where propargite was found to give excellent

control of *T. evansi* under laboratory and greenhouse conditions. Kithusi (2005) reported that propargite was effective in controlling *T. evansi* in the greenhouse and field conditions.

Greenhouse findings confirm the laboratory findings in that the leaf damage and mite populations were kept low by application of dicofol, profenofos+cypermethrin, propargite, bifenthrin and lambda-cyhalothrin. Dimethoate application slightly decreased mite populations compared to the control but the mite populations kept increasing regardless of the pesticide application. There was gradual increase in leaf damage in both the Dimethoate treated plants and the control.

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CHAPTER FOUR

EFFECT OF PESTICIDES ON GEOGRAPHICALLY ISOLATED FIELD POPULATIONS OF *Tetranychus evansi* IN KENYA

4.1 Abstract

Tobacco spider mites (*Tetranychus evansi*) collected from Loitoktok, Kibwezi, Athi-River and Subukia were evaluated for their susceptibility to bifenthrin, dimethoate, lambda-cyhalothrin, dicofol, propargite and profenofos+cypermethrin. Dicofol, bifenthrin, propargite and profenofos+cypermethrin caused significant mortality in the mites from all the populations tested, however, mortality did not differ significantly between the populations for each of these pesticides ($P<0.05$). Pooled data also showed that mortality did not differ signicantly between these acaricides with average contact adult mortalities of 100%, 99.67%, 99.83% and 99.83% respectively. Lambda-cyhalothrin caused significant mortality in mites from Kibwezi. However, mites from Loitoktok, Subukia and Athi-River showed significant tolerance. There were significant levels of tolerance to dimethoate in all the field populations treated with the highest adult mortality of 42.5% for Kibwezi population. Pooled data shows that there was significant difference ($P<0.05$) between lambda-cyhalothrin (65.33%), dimethoate (20.5%), the control (1%) and the rest of the chemicals tested. Mites from most Kenyan farms respond well to bifenthrin, propargite, profenofos+cypermethrin and dicofol. Dimethoate and lambda-cyhalothrin should be avoided

4.2 Introduction

Chemical control has been widely used to control the tobacco spider mites in Kenya and the use of these chemicals has continued to rely on the effectiveness of these chemicals in eliminating the pest (Mabeya *et al.*, 2002). However, continued exposure of mites to the same chemical over time leads to resistance (Cranham and Helle, 1985; Reissing *et al*., 1986). Due to the absence of an effective natural enemy in Africa yet and because few new acaricides are under development, and even fewer are acceptable in integrated pest management programs, it is important to both preserve and maximize the effectiveness of available products (Marshall and Pree, 1991).

Given the increasing pest status of *T. evansi* in Kenya and in Africa, there is need to develop control packages that are sustainable. It is therefore expedient that studies to determine the susceptibility of *T. evansi* to the commonly used acaricides and insecticides are carried out in order to gain information on the possible resistance by *T. evansi* to the pesticides, and as a result, only those pesticides that show continued toxicity are recommended for use by farmers. It is also important to determine if resistance is a problem in a specific location or time in order to improve pesticide choice (Dennehy and Granett, 1984). This will in turn reduce the possibility of pest resurgence associated with high use of pesticides.

4.3 Materials and methods.

4.3.1 Collection of mites from different locations

Red spider mites were collected from the following important tomato growing zones of Kenya: Mwea Division in Kirinyaga District, Loitoktok Division in Kajiado

District, Subukia Division in Nakuru District, Kibwezi Division in Makueni District, Mtwapa Division in Kilifi District and Athi-River Division in Machakos District. Information on control practices by the farmers was collected through direct interviews of farmers. The type of pesticides used plus the frequency of application were recorded (Appendix 2). The global positioning system (GPS) location of the areas was taken.

The sampling procedure entailed collecting 20-30 infested leaves per farm in all the farms where mites were seen. A total of 31 farms were sampled as follows: 4 farms in Loitoktok, 3 farms in Mtwapa, 9 farms in Mwea, 2 farms in Kibwezi, 6 farms in Bahati and 6 farms in Athi-River. Sampling was done between April and July 2004. There were differing numbers of farms under tomato production during this season hence the differences in the number of farms sampled per location.

The leaves were first examined to ensure they had mites using a hand lens (X10). The leaf samples were put in paper bags, then placed in a cool box and transported to the laboratory at ICIPE. In the laboratory, 30-50 adult female spider mites were sampled from the infested leaves collected from each field; no more than one spider mite was taken from any leaf. The spider mites were first identified to ensure that they were *T. evansi* before being multiplied.

4.3.2 Identification of spider mites from different locations.

In the laboratory, at least twenty adult males from each of the sites sampled were picked under a stereo microscope with a hair brush and transferred into small vials

containing 70% alcohol where they stayed for ten days for the purpose of clearing to remove the internal tissues (Craemer *et al.*, 1998).

The mites were then mounted for identification. A small drop of polyvinyl alcohol (PVA) was placed in the middle of a clean slide. A mite was then transferred to the drop using the hair brush and was manoeuvred to ensure it lay in a dorsoventral position. A cover slip was placed over the specimen by holding it on its edge on the side of the drop touching the PVA medium then gently lowering it onto the drop. The mounted slides were then allowed to dry in the oven at 30°C for 24 hours (Craemer *et al.*, 1998).

Identification was done under X400 power magnification of the microscope. The mites were identified up to species level using the shape of the male aedegus and the position of the duplex setae as the distinguishing characteristics as described by Craemer *et al*. (1998) and Meyer (1987).

4.3.3 Culturing the mites collected from the field in the laboratory

Red spider mites from Loitoktok, Subukia, Kibwezi and Athi-River were identified as *T. evansi* while from Mwea and Mtwapa were *T. urticae*. Only *T. evansi* was reared in water isolation cages designed after those described by Dennehy and Granett (1982) for laboratory bioassays. Potted tomato plants were placed on empty inverted pots inside a basin half-filled with water. The rectangular clear-sided 3mm thick cages made of perspex material, with openings on the top side covered with fine polyester lining for ventilation was placed over the potted plants such that the open basal end of the cage was submerged in water. The spider mites were reared on 4-6 week old tomatoes (commercial variety Cal-J) under room temperatures and humidity in the laboratory.

4.3.4 Ovicidal tests

Using the mites isolated from different regions with the laboratory culture of *T. evansi* as a control, for each pesticide, twenty *T. evansi* females from the laboratory colony were transferred to tomato leaf discs (25mm) placed on moistened cotton wool in petri dishes. The females were allowed to oviposit for 24 hours after which the mites were removed and the number of eggs adjusted to twenty. Pesticide solutions were prepared using the manufacturer's recommended rates for each pesticide as earlier described in 3.3.1.3

The leaf discs containing the eggs were dipped in the pesticide solutions for five seconds (Keena *et al.*, 1991). Control discs were placed in water containing aquawett sticker. The treatments were replicated six times. The treated leaf discs were placed upside down in plastic petri dishes containing moistened cotton wool. Saturated solutions of potassium carbonate were used to maintain humidity conditions (Winston and Bates, 1960) in the plastic boxes (22 by 33 by 7cm) in which the trays containing the petri dishes were placed. The boxes were covered and kept in the incubator at 25^0C. The experiment was arranged in a complete randomized design (CRD).

The leaves were examined after 4 days for hatched larvae and were further examined daily for another five days. Egg mortality was determined by comparing the number of unhatched eggs with the original number of post treatment eggs (Agnello *et al.*, 1994).

4.3.5 Adulticidal tests.

Two bioassay methods modified leaf direct method (LDD) and leaf disc residue – dipping (LDR-D) were used in this study using the mites from each of the six

locations. The two methods are a modification of those described by Kabir *et al.*, (1993)

In the modified leaf disc direct method, chemical solutions were prepared using the manufacturer's recommended rates as described in 3.3.1.3 above. Twenty same age adult females (Knowles *et al.*, 1988) of *T. evansi* from the laboratory culture were transferred onto the lower side of 25mm diameter tomato leaf discs using a fine brush. The leaf discs containing the mites were dipped in the pesticide solutions for 5 seconds and the discs placed upside down in plastic petri dishes containing moistened cotton wool. This was done for the six pesticides and water plus sticker as control with six replications each. The petri dishes were left uncovered for half an hour to allow the leaves to dry. Saturated solutions of potassium carbonate were used to maintain humidity conditions(Winston and Bates, 1960) in plastic boxes (22 by 33 by 7cm) in which the trays containing the petri dishes will be placed. The boxes were covered and kept in the incubator at 25^0C. The treatments were arranged in a completely randomized design (CRD).

The mites were observed after 24 hours and scored as dead or escaped for those which were trapped in the cotton barrier (Agnello *et al.*, 1994).

For the leaf disc residue – dipping method procedure in LDD method was repeated with the exception that the leaf discs were dipped first in the pesticide concentrations for 5 seconds. They were placed in petri dishes and allowed to dry at room temperature. The mites were then transferred onto the discs. The mites were then

observed after 24 hours and scored as dead or escaped for those which were trapped in the cotton wool (Agnello *et al.*, 1994).

The data was statistically analysed using ANOVA and means separated using Student-Newman-Keuls (SNK) test (SAS, 1990). The data was arcsin transformed before analysis but mean values in the table are actual percentage values. Correction for control mortality was done where mortalities in the control were > 20% using Abbott's formula (1925):

Pt = Po-Pc/ 100-Pc x 100,

where Pt = corrected mortality, Po = observed mortality and Pc = control mortality (all %s).

4.4 Results

4.4.1 Incidence of spider mites on tomatoes and use of pesticides in different production areas

Most farmers planted Cal J variety of tomatoes (Table 5). From the farms sampled, propargite is the most commonly used pesticide with 29% of the farms sampled using it. Profenofos+cypermethrin and dimethoate are the second with 22.6% farms using it. Dicofol and lambda-cyhalothrin were applied in 9.7% farms each. Bifenthrin and alpha-cypermethrin were used in 6.5% farms.

Generally, farmers from Subukia use profenofos+cypermethrin, those in Athi-River and Kibwezi use propargite and those in Loitoktok use dimethoate predominantly (Table 6). The farmers in Subukia complained of spider mites being a persistent problem in the area and that profenofos+cypermethrin was very effective for them because it controlled all the insects together with the mites satisfactorily. In Kibwezi

and Athi River, red spider mites were not ranked as being too problematic but the farmers interviewed had occasionally used the pesticides conventionally. It also appeared that most farmers in Loitoktok had a poor knowledge of *T. evansi* as an important pest of tomatoes and hence they used dimethoate as an insecticide to control all the pest problems they had.

Tetranychus evansi was found in Loitoktok, Kibwezi, Subukia and Athi-River. However, only *T. urticae* was collected in Mwea and Mtwapa (Table 7).

Table 5: Percentage of farmers growing different varieties in different tomato growing regions.

	Cal-J	Onyx	Rio-Grande
Loitoktok	50	25	25
Mtwapa	33.3	33.3	0
Mwea	66.7	44.4	0
Kibwezi	100	0	0
Subukia	66.7	16.7	16.7
Athi-River	100	0	0

Table 6: Percentage of farmers using different pesticides for spider mites control.

	Dicofol	Dimethoate	Bifenthrin	Propargite	Lambda-cyhalothrin	Profenofos + cypermethrin
Loitoktok	0	75	0	0	0	25
Mtwapa	0	33.3	33.3	0	66.7	33.3
Mwea	22.2	22.2	0	22.2	11.1	22.2
Kibwezi	100	50	0	0	0	0
Subukia	50	0	0	16.7	0	0
Athi-River	16.7	16.7	0	66.7	0	0

Table 7: Percentage of farms where different mites species were collected from the different tomato growing areas

	Tetranychus evansi	*Tetranychus urticae*
Loitoktok	100	0
Mtwapa	0	100
Mwea	0	100
Kibwezi	100	0
Subukia	83.3	16.7
Athi-River	100	0

4.4.2 Efficacy of pesticides on tobacco spider mites collected from different tomato producing areas.

There were no significant differences in egg mortality caused by the pesticides between field populations and the laboratory culture of mites maintained free of pesticides for three years at ICIPE (Table 8). In Loitoktok, there was significant tolerance to lambda-cyhalothrin both due to contact and residual effect of the chemical (Tables 9 and 10). Dicofol gave 100% ovicidal and adulticidal mortality both contact and residual. This pesticide is not being used by any of the farmers where samples were collected. There were significant differences in adult mortality between populations due to the residual effect of most of the chemicals. Pooled data results indicate that there was significantly high mite mortalities due to dicofol, propargite, bifenthrin and profenofos+cypermethrin generally with adult mortalities being more than 99% ($P<0.05$). There was a low level of tolerance to lambda-cyhalothrin and a higher tolerance to dimethoate adult mortalities of 65% and 25% ($P<0.05$) respectively (Table 12).

Table 8: The contact effect of the pesticides on the egg emergence of *Tetranychus evansi* eggs from mites from different tomato growing regions six days after treatment.

Pesticide	Egg mortality (%) ± SE				
	Athi-River	Loitoktok	ICIPE	Subukia	Kibwezi
Dicofol	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a
Bifenthrin	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a
Lambda-cyhalothrin	97.7±2.3a	99.0±1.0a	100.0 ±0.0a	95.6±4.4a	96.7±2.1a
Propargite	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a
Profenofos+cypermethrin	100.0 ±0.0a	100.0 ±0.0a	100.0 ±0.0a	95.6 ±0.0a	100.0 ±0.0a
Dimethoate	44.1±9.9b	39.5±8.9b	47.8±20.8b	58.8.2±7.7b	74.0±16.6b
Control	0.0±0.0c	0.00±0.0c	0.0±0.0c	0.0±0.0c	0.0±0.0c

Means followed by the same lower case letter within column are not significantly different (SNK-test, P=0.05). Population means followed by the same upper case letter within rows are not significantly different (SNK-test, P=0.05).

Table 9: The contact effect of the pesticides on *T. evansi* adult females from different tomato growing regions after 24 hours.

Pesticide	Adult mortality (%) ± SE				
	AthiRiver	Loitoktok	ICIPE	Subukia	Kibwezi
Control	1.7±1.1cA	0.8±0.8dA	0.0±0.0cA	0.0±0.0dA	2.5±1.1dA
Dimethoate	7.5±4.2cB	24.2±9.8cAB	3.33±1.7cB	21.7±5.6cAB	45.8±14.9cA
Lambda-cyhalothrin	60.8±6.9bAB	42.5±7.2bB	80.8±4.0bA	66.7±8.1bAB	75.8±7.4bA
Propargite	100.0±0.0aA	100.0±0.0aA	99.2±0.8aA	100.0±0.0aA	100.0±0.0aA
Profenofos+ cypermethrin	99.2±0.8aA	100.0±0.0aA	100.0±0.0aA	100.0±0.0aA	100.0±0.0aA
Dicofol	100.0±0.0aA	100.0±0.0aA	100.0±0.0aA	100.0±0.0aA	100.0±0.0aA
Bifenthrin	100.0±0.0aA	98.3±1.7aA	100.0±0.0aA	100.0±0.0aA	100.0±0.0Aa

Means followed by the same lower case letter within column are not significantly different (SNK-test, P=0.05). Population means followed by the same upper case letter within rows are not significantly different (SNK-test, P=0.05).

Table 10: The residual effects of the pesticides on *Tetranychus evansi* adult females from different tomato growing regions after 24hours.

Pesticide	Adult mortality (%) ± SE				
	Athi-River	Loitoktok	ICIPE	Kibwezi	Subukia
Kelthane	100.0±0.0aA	100.0±0.0aA	100.0±0.0aA	100.0±0.0aA	95.8±3.3aA
Polytrin	93.3±4.4aA	66.7±12.6bAB	56.7±12.7bB	43.3±9.2bB	52.5±5.0bB
Omite	85.0±6.2aA	86.7±6.5aA	84.2±3.0aA	51.7±13.5bB	37.5±2.1cB
Brigade	54.2±9.8bAB	61.7±3.1bA	36.7±8.2cBC	20.0±5.2cC	32.5±3.6cBC
Karate	50.8±13.4bB	29.2±5.5cB	80.8±4.4aA	5.0±3.4cC	44.2±8.3bcB
Dimethoate	4.2±2.4cA	1.7±1.1dA	0.0±0.0dA	0.0±0.0cA	4.2±1.5dA
Control	2.5±1.1cA	2.5±1.7dA	0.0±0.0dA	0.0±0.0cA	2.5±1.7dA

Means followed by the same lower case letter within column are not significantly different (SNK-test, P=0.05). Population means followed by the same upper case letter within rows are not significantly different (SNK-test, P=0.05).

Table 11: **Pooled average contact effects of the pesticides on the eggs and adult females of *Tetranychus evansi* from different tomato growing regions.**

Pesticide	Adult mortality(%) ± SE	Egg mortality(%) ± SE
Dicofol (2.5ml/l)	100.00±0.00a	100.0±0.0a
Profenofos+cypermethrin (1.5ml/l)	99.83±0.17a	100.0±0.0a
Propargite(1ml/l)	99.83±0.17a	100.0±0.0a
Bifenthrin (2.5ml/l)	99.67±0.33a	100.0±0.0a
Lambda-cyhalothrin(2.5ml/l)	65.33±3.77b	97.8±1.1a
Dimethoate (1ml/l)	25.50±4.51c	54.3±5.9b
Control (water+sticker)	1.00±0.37d	0.0±0.0c

Means followed by the same letter are not significantly different (SNK-test, P=0.05). Percent mortality data were arcsin-transformed before analysis but mean values in the table represent actual percentage of mortality.

4.5 Discussion

Tetranychus evansi continues to gain importance given that from the samples identified, it was found to be the predominant species in Loitoktok unlike the findings by Machini (2005) from surveys done in 2002 that showed that *Tetranychus urticae* was the predominant species. It was also identified in Kibwezi and Athi-river areas, these being areas where *T. evansi* have not been reported before. The most commonly used pesticides by the farmers are propargite, profenofos+cypermethrin, dimethoate and lambda-cyhalothrin. It is evident that most of these pesticides are still effective against *T. evansi* both as ovicides and adulticides. Dicofol is specific and the most effective acaricide. All the mites treated with this pesticide were effectively controlled. It however was not used by any of the farmers where mite samples were collected. Propargite, profenofos+cypermethrin and bifenthrin are highly effective. This differs with farmer complaints of poor control with propargite in the fields which can be attributed to poor application techniques. There are chances that with time, tolerance to lambda-cyhalothrin may be expected in Loitoktok where in as much as the farmers interviewed did not use lambda-cyhalothrin for spider mite control, the mites sampled were slightly tolerant to it showing that the farmers could have been using it to control other pests resulting in tolerance in a non-target pest. Tolerance to Dimethoate was recorded in all the mite populations tested. This tolerance is widespread and often at a high level even up to 1000-fold in Zimbabwe (Blair, 1989).

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CHAPTER FIVE

GENERAL DISCUSSION, CONCLUSIONS AND RECOMMENDATIONS

Laboratory and greenhouse evaluation confirm that the manufacturer's recommended rates of dicofol, bifenthrin, profenofos+cypermethrin, propargite and lambda-cyhalothrin for the control of mites are effective in controlling *Tetranychus evansi*. However, dimethoate is weak as an acaricide against *Tetranychus evansi.* This agrees with findings by Blair (1989) in Zimbabwe that dimethoate did not effectively control mites.

It is evident that *T. evansi* is gaining importance given its appearance in more sites like Kibwezi and Athi-river where it was not reported before. It is also gaining importance in places where it was recently identified like the case of Loitoktok where in a survey carried out in 2002 by Machini (2005), *T. urticae* was the predominant species whereas from this study *T. evansi* is now the predominant species. There is a possibility that it is spreading to other parts too due to human travel and product transfer to further markets.

There is still a wide range of chemicals that can be used effectively in rotations to control *T. evansi* in tomato production in Kenya. It is clear that bifenthrin, propargite, profenofos+cypermethrin and dicofol can still be used effectively for *T. evansi* control. There are signs that mites may develop tolerance to lambda-cyhalothrin which despite the fact that it is not registered as an acaricide, farmers have been using with considerable level of success in some areas and laboratory results also show its toxicity to the tobacco spider mite. This can be attributed to the fact that farmers use

this pesticide to control other insect pests. Dimethoate is among the most used acaricide by the farmers and experiments indicate that there is a high level of tolerance to it.

Farmers' complaints of poor chemical control could be attributed to lower rates of application, longer spraying intervals and poor spraying techniques due to lack of understanding of the pest behaviour. Most farmers do not observe any preventive measures in that they keep transferring stakes from infested fields to other fields hence increasing sources of infestations. Other factors that could lead to inability of the pesticides to effectively control the mites is the web developed by the mites that act as a protective canopy holding spray droplets and that is the reason why Davis (1952) and Linke (1953) urgently advocated the destruction of webs during chemical control procedures. The database of Arthropods resistance to pesticides does not list *T. evansi* (Whalon & Mota-Sanchez, 2000) as one of the pests which have developed resistance.

From this study, the following recommendations can be made:

- Dimethoate should not be recommended as a pesticide for *T. evansi* control.
- Pesticide mixtures like profenofos+cypermethrin should be encouraged for use by farmers. Other effective pesticides like propargite, bifenthrin and dicofol can be used in rotations.
- Use of lambda-cyhalothrin should be avoided because of its tendency to cause tolerance.

- A more extensive study should be done to determine the pest status of *T. evansi* in Kenya.
- Farmers should be trained on pest behaviour, proper spraying techniques and preventive measures like destruction of previously infested crop residues.
- Further work should be done under field conditions to determine how many exposures of the same pesticide would lead to development of resistance.

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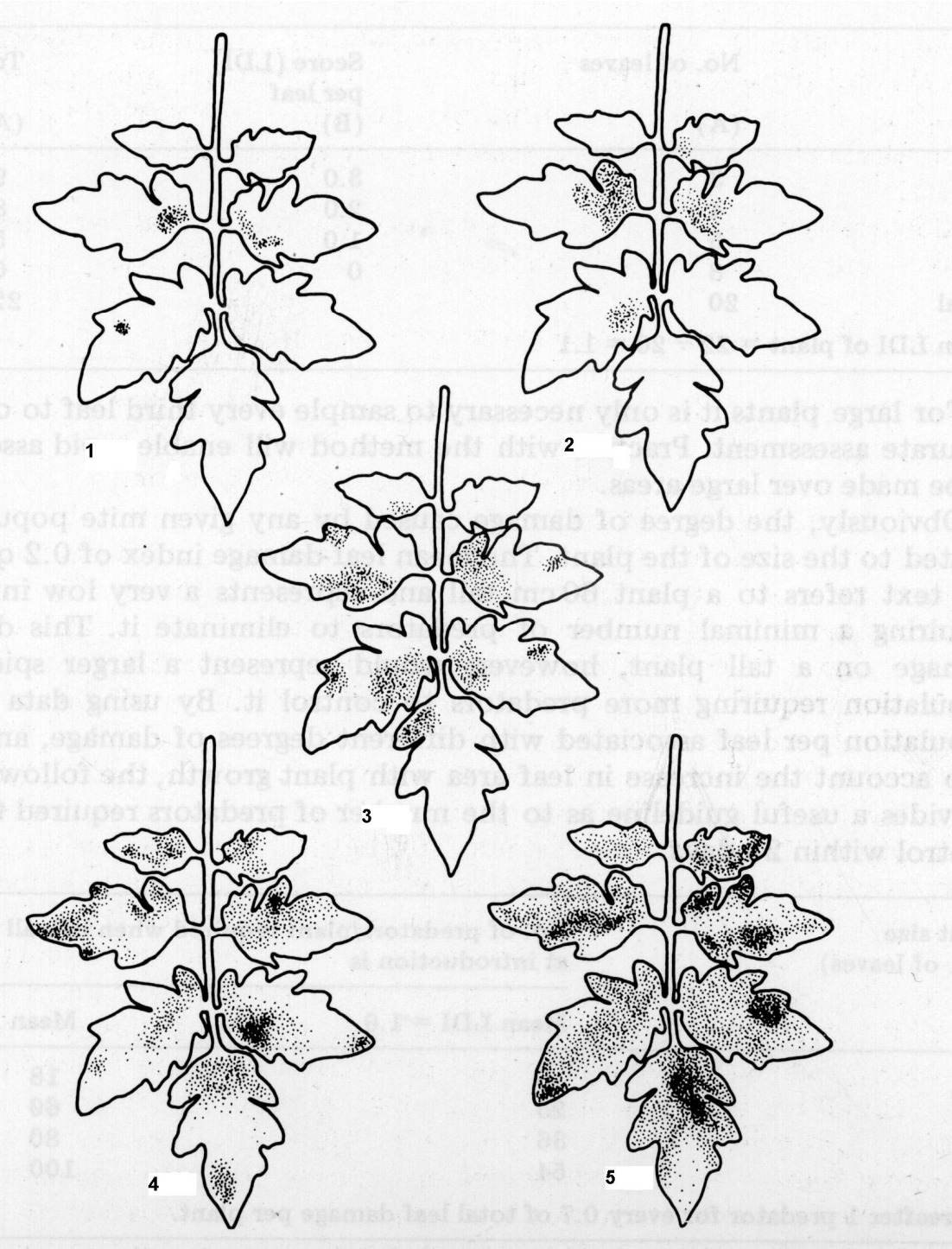
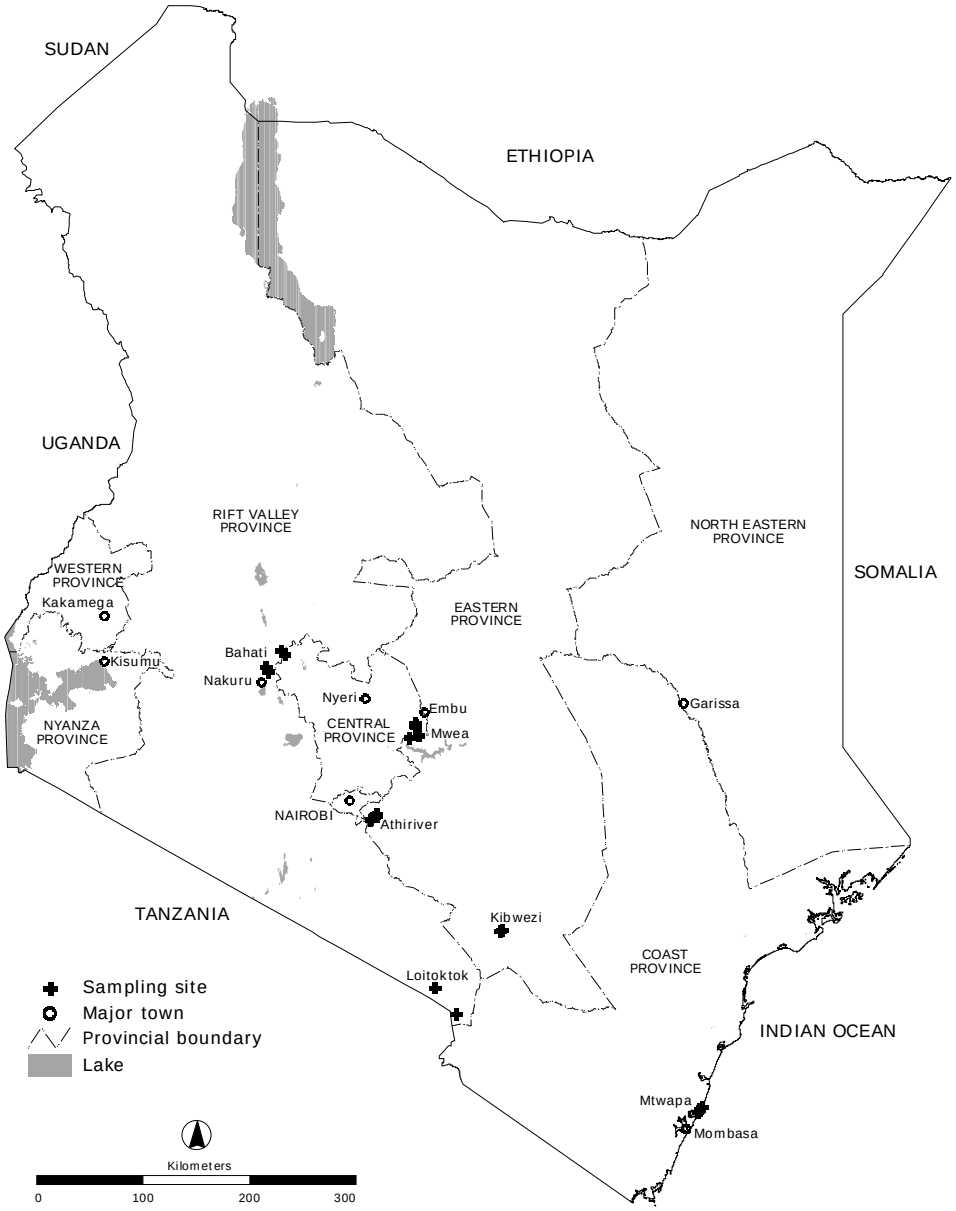
APPENDICES:

Appendix 1: Damage score on a scale of 1 – 5 (Hussey and Scopes)



Appendix 2: A map showing the geographical location of the sites where the mites used were sampled from



Appendix 3: Pesticide resistance field questionnaire

District: …….. Division: ……

Farmer's name: ……. Time: …….. GPS: ……..

Size of field under tomatoes:…....……..

Varieties planted: 1. …….. 3. ……

2……....

Pesticides used:

Pesticide	**Frequency of application**

Comments:……..
……
……
……
……
……
……

Appendix 4: The Geographical position co-ordinates of farms where tomatoes were sampled from

AREA	FARM	GPS	
Loitoktok	1	02°21.57S	038°06.3E
Loitoktok	2	02°51.20S	037°32.11E
Loitoktok	3	02°51.20S	037°32.11E
Loitoktok	4	03°04.24S	037°42.54E
Mtwapa	1	03°54.07S	039°45.30E
Mtwapa	2	03°51.27S	039°47.45E
Mtwapa	3	03°52.47S	039°46.30E
Mwea	1	00°38.29S	037°22.25E
Mwea	2	00°38.10S	037°22.16E
Mwea	3	00°37.14S	037°22.01E
Mwea	4	00°38.42S	037°22.36E
Mwea	5	00°38.18S	037°21.58E
Mwea	6	00°44.18S	037°18.75E
Mwea	7	00°44.18S	037°18.75E
Mwea	8	00°44.49S	037°18.70E
Mwea	9	00°42.90S	037°23.58E
Kibwezi	1	02°21.54S	038°06.15E
Kibwezi	2	02°22.25S	038°05.17E
Subukia	1	00°02.34S	036°15.62E
Subukia	2	00°00.27S	036°13.87E
Subukia	3	00°10.76S	036°06.96E
Subukia	4	00°10.76S	036°06.96E
Subukia	5	00°08.49S	036°05.87E
Subukia	6	00°08.57S	036°05.93E
Athi River	1	01°23.50S	037°02.43E
Athi River	2	01°23.03S	037°02.29E
Athi River	3	01°26.15S	036°59.36'E
Athi River	4	01°25.51S	036°59.51E
Athi River	5	01°23.38S	037°02.05E
Athi River	6	01°24.29S	037°01.18E