# Resistance status of the tobacco spider mite (*Tetranychus evansi*) to currently recommended acaricides on tomatoes (*Lycopersicon lycopersicum*) and the effects of these acaricides on natural enemies of vegetable pests



By

**Rowena Judith Kwaramba**

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Department of Crop Science
Faculty of Agriculture
University of Zimbabwe

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# Abstract

Laboratory studies were performed on tobacco spider mites (*Tetranychus evansi*) collected from Chinamhora and Mutoko to investigate grower claims of resistance to commonly used acaricides. Bioassays of *T. evansi* collected from tomatoes showed presence of resistance to dimethotae, malathion, abamectin and amitraz. The RF values for Chinamhora were 1.2 for abamectin,1.0 for malathion and 1.4 for amitraz and dimethoate. For Mutoko RF values were 1.2 for abamectin and malathion, 1.4 for amitraz while it was 12.1 for dimethoate. The farmers could have used these chemicals for a long time without changing them especially dimethoate. In addition, their water quality could have been compromising control, as pesticides are effective with clean water. In the field, there was no evidence of resistance although populations increased as the season progressed. Damage leaf indices for site one ranged from 0-3 for the 4 weeks while those of site two were between 0-4 in the 7 weeks data was collected. At both sites, malathion and dimethoate had the highest indices showing that they were not as effective as the other acaricides. Dimethoate however performed better than malathion. From a visual assessment, abamectin was the most effective followed by amitraz. Using the risk category for the natural enemies, bioassays showed that all the acaricides were harmful. Malathion and dimethoate were slightly harmful to the ladybird beetle but moderately harmful to the other three natural enemies. Amitraz was moderately harmful to all natural enemies. Abamectin was moderately harmful to the ladybird beetle and aphid wasp parasitoid while harmful to the predatory mite. As a result of this experiment control options and recommendations as part of an insecticide resistance management strategy were discussed briefly. These, it is envisaged, would delay or slow down development of insecticides resistance (IR).

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# Dedication

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# CHAPTER ONE

# INTRODUCTION

## 1.1. Background

Tomato (*Lycopersicon lycopersicum*) is one of the most important vegetables grown in Zimbabwe. It ranks either first or second to leaf vegetables depending on location. It is an important source of vitamins and an important cash crop in both the smallholder and commercial farming sectors (Turner and Chivinge, 1999). In the smallholder sector yields have remained very low due to poor management especially pest control. One of the major pests is the tobacco spider mite (*Tetranychus evansi*) (Kochhar, 1986). Currently spider mite control involves weekly applications of acaricides (Dagnoko and Kwaramba, 2000). However, the practice has detrimental effects on pests, natural enemies, human beings and the environment. In Zimbabwe farmers are having problems controlling mites with the available chemicals.

Insecticide resistance results from the selection of insect strains tolerant to doses of insecticide that would kill the majority of the normal population (Cremlyn, 1978). Such strains are rare in a normal population, but the widespread use of an insecticide can reduce the susceptible individuals providing the resistant strain with a competitive advantage. The resistant individuals multiply in the absence of intra-specific competition, quickly becoming dominant. Insecticide resistance develops fast under shorter life cycles, little immigration of susceptible individuals and increased intensity of pesticide use, which is very typical of red spider mites (Pedigo, 1996). Resistance impairs chemical

control due to selection of resistance genes leading to decreased control efficacy. Currently it is very difficult and expensive to develop and register new compounds, and the sequential selection of resistance mechanisms broadens the overall cross-resistance spectrum predisposing pests to resist new, yet unused pesticides (Denholm, 1992). Insecticide resistance development has led to the rationalisation of pesticide use and development of alternative strategies for conserving the effectiveness of existing and new chemicals. Emphasis is now shifting to more rational use of insecticides to preserve their long-term effectiveness. This approach is known as insecticide resistance management (IRM).

In nature, natural enemies regulate pest population numbers returning them towards equilibrium and restricting the numbers within limits (Mark and Kidd, 1996; Verkerk, 2001). Predatory mites, *Phytoseiulus spp* and *Amblyseius spp*, are effective predators of the phytophagous mites. They attack mites in areas of the plant difficult to target with contact pesticides (Matthews, 1984). Generally, pesticides are believed to have lethal and sub-lethal effects on these natural enemies. They may be killed directly following exposure by contact, ingestion or by respiration. Natural enemies are usually more susceptible to pesticides owing to their general small size, searching habits, usually less-developed enzyme-based detoxification systems and preening behaviour (notable in parasitoids) (Verkerk, 2001). Indirectly, natural enemies are killed when hosts are killed. Not much work has been done in Zimbabwe concerning the effects of pesticides on natural enemies.

The aim of this research was to investigate the resistance status of the tobacco spider mite (*T. evansi*) to currently recommended and mostly used acaricides in Zimbabwe. Resistance has been reported in spider mites in cotton (Duncombe, 1972; Brettell, 1995). The chances are high that the tobacco spider mite (*T. evansi)* could develop resistance since the same acaricides are used in tomatoes. Investigating resistance is crucial to the development and implementation of an insecticide resistance management (IRM) strategy to suit local agricultural needs. The research was also seeking to find out if these acaricides had any effects on major natural enemies of vegetable insect pests. This knowledge is vital in the implementation of integrated pest management (IPM) strategies.

## 1.2. Objectives

The objectives of this research therefore were:

1. To investigate the resistance status of the tobacco spider mites (*T. evansi*) to the currently recommended and mostly used acaricides.
2. To investigate the effects of currently recommended and mostly used acaricides on natural enemies of red spider mites.

## 1.3. Hypotheses

The hypotheses were that:

1. The tobacco spider mite has developed resistance to currently recommended and mostly used acaricides.
2. The currently recommended and mostly used acaricides have negative effects on natural enemies.

# CHAPTER TWO

# LITERATURE REVIEW

## 2.1. Tomato production in Zimbabwe

Tomato (*Lycopersicon lycopersicum*) is a subtropical plant belonging to the Solanaceae family together with the Irish potato, eggplant, paprika and the peppers. A native of Ecuador and Peru, it was introduced to Europe in the 16$^{th}$ century and was listed as an ornamental because of its relationship with the deadly black nightshade (*Solanum nigrum*) (Kochhar, 1981). It was only accepted as edible in the 1800s and is one of the most important vegetables grown and consumed world wide today. Tomato is a perennial plant grown as an annual and includes determinate (bush), semi-determinate and indeterminate (vining) types. It is grown mainly for fresh fruit consumption and for cooking. It has also become an important raw material in the processing industry, and for research into fundamental principles of growth and development in plants. It is generally ranked high amongst the long list of vegetables grown in Southern Africa (Turner and Chivinge, 1999). In Zimbabwe, it ranks either first or second to leaf vegetables depending on the farming area. Tomatoes are grown at all levels from the backyard garden of almost every homestead up to large hectarages in sub-Saharan Africa. The fruit contains 94% water, 1% protein, 4% carbohydrates and significant amounts of vitamin A (20%), $B_2$, C (ascorbic acid), potassium, dietary fibre, calcium, iron, thiamine, nicotinamide and magnesium (Wells, 1980; Kochhar, 1986).

Yields have remained far below the crop's potential in the smallholder sector due to poor quality seeds, the general non-availability of horticultural inputs, poor management and the large numbers of pests that attack tomatoes (GTZ, 2000). In the smallholder sector yields as low as 7t/ha were reported in Tanzania, 10t/ha in Uganda and 12t/ha in Zimbabwe compared to at least 100t/ha in Zimbabwe in the commercial sector (Varela and Seif, 2000). Pests of tomatoes include the blight, wilt, viral and canker diseases, nematodes and several arthropod pests. Amongst the arthropod pests, tobacco spider mites (*Tetranychus evansi*) are currently the most damaging dry season, non-insect arthropod pests of tomato in the semi-arid areas (Varela and Seif, 2000). These are then followed by insect pests namely the African bollworm (*Helicorvepa armigera*), whiteflies (*Bemisia tabaci*), several leafminer species, thrips (*Thrips tabaci*) and tomato russet mites or tomato rust mites (*Aculops lycopersici* (Tryon)) an arthropod pest (Varela and Seif, 2000; Verkerk, 2001). Tobacco spider mites are a major pest of an important crop, the tomato, and so their resistance status to currently recommended and mostly used acaricide is worth establishing.

## 2.2 Tobacco spider mites

### 2.2.1 Distribution and pest status

Tobacco spider mites are widely distributed throughout the world (GTZ, 2000; Pedigo 1996). They have been reported to attack tomatoes in temperate regions under protected environments and can multiply rapidly in greenhouses (Varela and Seif, 2000). Mites attack crops in open fields in warmer areas. The tobacco spider mites have an extremely wide host range from a variety of cultivated crops as well as wild plants. They are

common on tomatoes, tobacco, potatoes, soya beans and beans, sunflowers, groundnuts, green peppers, citrus and deciduous fruits, strawberries, cucurbits, and ornamentals. They seem unselective often moving on to adjacent plants when their original hosts become overpopulated or have been sprayed (Taylor, 1981). Several *Tetranychus* species occur worldwide. The most important species on tomatoes in Southern Africa (Malawi, Mozambique, Namibia, South Africa, Zambia and Zimbabwe) is *T. evansi* Baker and Pritchard (the tobacco spider mite), *T. urticae* Koch (the two-spotted spider mite) and *T. cinnabarinus* (Boisduval) (the carmine spider mite or common red spider mite) (GTZ, 2000). Other *Tetranychus* species infesting tomatoes, but of minor importance, include *T. lombardinii* Baker and Pritchard (the crimson spider mite), *T. ludeni* Zacher (the dark red mite or red-legged spider mite), *T. neocaledonicus* Andre' (the vegetable spider mite) and *Eutetranychus orientalis* (Klein) (the oriental red mite). Red spider mite is the term most commonly used to describe the economically important *Tetranychid* mites. According to Duncombe (1972), *T. cinnabarinus, T. lombardinii* and *T. ludeni* were more common in cotton than in tomatoes.

*T. evansi* was accidentally introduced to Southern Africa from South America (Brazil) in the early 1980s, but has become the most important dry season arthropod pest of tomato in Southern Africa today (Kochhar, 1986, Varela and Seif, 2000). It favours Solanaceous crops namely the tomato, Irish potato, eggplant, tobacco, and related weeds. Some of the weeds which host the tobacco spider mites acting as reservoirs of the pests include bobbin weed (*Leucas martinicensis),* upright starbur *(Acanthospermum hispidum),* pigweed (*Amaranthus hybridus*), Sabi morning glory (*Ipomoea plebeia),* apple of Peru

(*Nicandra physaloides*), black nightshade (*Solanum nigrum*), bitter apple (*Solanum spp*), wild gooseberry (*Physalis angulata*) and stinkblaar (*Datura stramonium*). The tobacco spider mite is most severe on tomato when persistent dry conditions coupled with high temperatures of 16°C- 37°C are persistent, which results in increased developmental rates of the newly born nymphs. Widespread rains and high humidity suppress populations. They have however been known to attack all year round even in the nurseries. Mites are spread by wind over long distances. Animal movements, birds and people aid dispersal. They can be spread passively by irrigation water, dust storms, clothing and implements (Duncombe, 1976; Wells, 1980; Grout *et al*, 1998).

Spider mites have not always been important pests in Zimbabwe. It happened only after the introduction of a bollworm control programme using carbaryl and DDT in cotton in the 1960s (Brettell, 1995). They have been described as a pesticide-induced pest by several authorities (Duncombe, 1972; Brettell, 1995; Verkerk, 2001). Only after insecticide and/or acaricides applications with resultant predator destruction do mites become a real problem (Grout *et al*, 1998). Some insecticides like the pyrethroids such as cypermethrin and deltamethrin have been reported to enhance red spider mite reproduction (Varela and Seif, 2000). Verkerk (2001) stressed that most serious spider mite problems are on crops that are sprayed regularly with acaricides or insecticides. According to Duncombe (1972), use of DDT in cotton resulted in improved plant vigour and growth, promoting higher nitrogen and sugar contents resulting in positively correlated mite populations. DDT was also believed to stimulate female mites to deposit greater numbers of eggs (Duncombe, 1972). He also hypothesised that the DDT-treated

plants and surfaces led to mite dispersal due to irritation and repellency resulting in less intra-specific competition hence more food per individual and greater rate of reproduction. Peak populations were then reached earlier. Duncombe (1972), believed that the inert residues (for example talc) used in some DDT formulations made the leaf stratum more suitable for the adherence of mites and thus promoted population increases. Brettell and Burgess (1971) supported the same argument when they reported high mite populations on plants adjacent to dusty roads.

### 2.2.2. Description

Under the Kingdom Animalia and Phylum Arthropoda, mites belong to the class Arachnida and order Acarina. They belong to the super family Tetranychoidea, family Tetranychidae and genus *Tetranychus* with several species. These are very tiny animals of <1mm long with eight legs, except the larvae that have six. The different mite species are very difficult to distinguish without a microscope. Plate 2.1 indicates major differences of the important female *Tetranychid* species while Plate 2.2 shows the tomato russet mite (*Aculops lycopersici*) (Magdalena & Meyer 1981).

**A** **B** **C** **D** **E**

**Plate 2.1** Females of: *Tetranychus cinnabarinus* **(A)** *T. urticae* **(B);** *T. evansi* **(C),** *T. lombardinii* **(D)** and *T. ludeni* **(E)** (*Pictures* **A, B, D,** *and* **E** *courtesy of Magdalena 1981;* **C** *courtesy of Varela and Seif 2000*)

**Plate 2.2** *Aculops lycopersici* (Tomato Russet mite)
(*Picture courtesy of Magdalena 1981)*

The females continuously secrete and spin a network of very thin silk threads producing fine webbing (Plate 2.4). This forms a fine protective covering above the mite colonies, over which they move across the leaf surface and anchor themselves and their eggs to the plant. This web also protects them from enemies and pesticides (Verkerk, 2001). *T. evansi* produces more silk than any other species and in high infestations leaves are completely covered by the web.

### 2.2.3. Life cycle of the tobacco spider mite

Tobacco spider mites have a life span of 13 - 32 days. Adult females can lie dormant for a long time and over-winter in underground litter, tree barks or crop residues before colonizing plants. Most species over-winter in the egg stage. They become active in early spring and oviposit. Each female lays between 10 - 15 eggs a day and an average of 200 eggs in its life. Eggs hatch in 3 - 10 days. Mites undergo incomplete metamorphosis with 3 post-embryonic and quiescent stages that include the six legged first instar larvae which moults into the second eight-legged instar larvae (protonymph), then the third instar (deutonymph) stages before becoming adults (Pedigo, 1996). From egg to adult, they also undergo 3 quiescent stages i.e after hatching, first and second instars. In summer, it takes 10 - 13 days from egg to adult. Females may live up to 9 weeks. There are numerous generations per year with population peaks occurring in September and October in Zimbabwe. A single female mite is capable of initiating a new infestation as sex is determined by fertilization. Males hatch from unfertilized eggs while females hatch from fertilized eggs (Varela and Seif, 2000, Borror *et al*, 1984).

### 2.2.4. Damage

The tobacco spider mites, like most spider mites, normally prefer to inhabit the underside of leaves but in severe infestations, they will occur on both surfaces, stems and fruits. The first visible sign of plant damage is a characteristic small silvery-yellow mottling on the upper leaf surface especially between the main veins, near the leaf stalk (Plate 2.3). Mites tend to aggregate on the tips in high infestations (Plate 2.4). Later, affected areas spread. Attacked leaves turn dull dirty-grey on the upper surfaces, chlorotic to bronze coloured then brownish with thin mosaic patterns on the underside and eventually fall off. All active stages suck sap from tomato leaves, stems and fruits. According to Pedigo, (1996) such feeding destroys plant cells reducing the photosynthetic area leading to low yields and fruit quality. Tomato quality from the consumers' point of view consists of attributes such as appearance, flavour, shelf life and nutritional composition. Defoliation by mites leads to production of smaller and lighter fruits with a low percentage of soluble solids and a lower content of ascorbic acid (Varela and Seif, 2000). The fruits fail to get the normal red colour at ripening, but whitish speckling patches and spots appear on the skin (Plate 2.5A and B). The whole plant may become covered in webbing. The female mites and the webbing are visible to the naked eye (Plate 2.4). The pest can actually wipe out a whole field when a crop is attacked in its early stages and no control measures are taken. The damage caused by the russet mite is shown in Plate 2.6.

Plate 2.3

Plate 2.4

**Plate 2.3.** Typical tobacco spider mite damage on tomato leaf (C*ourtesy of Dobson et al, 2002)*

**Plate 2.4.** High infestation of tobacco spider mites on the tip of a tomato leaf (note the web)

A

B

**Plate 2.5** Tobacco spider mite damage on unripe (A)(C*ourtesy of Dobson et al, 2002)* and ripe fruits (B) (C*ourtesy of Varela and Seif, 2000*)

**Plate 2.6** Tomato leaf damage by tomato russet mite *(Courtesy of Magdalena & Meyer, 1981)*

### 2.2.5. Current control practices

During the dry season tomatoes cannot be grown profitably without continuous control of arthropod pests, mainly the tobacco spider mites. Current control has been based on weekly applications of acaricides and insecticides (Varela and Seif, 2000). Several surveys conducted and observations made in Zimbabwe established use of chemicals like dimethoate (Rogor), amitraz (Mitac), malathion (Malathion) and dementon-S-methyl (Metasystox) in the smallholder sector (Dagnoko and Kwaramba, 1999). Many more acaricides are registered for red spider mite control in tomatoes in Zimbabwe (Table 2.1).

Spraying is usually effective at an early stage of infestation. Other insecticides recommended for deciduous fruit trees and ornamentals, which include monochrotophos and triazophos, are sometimes used on tomatoes. According to Matthews (1984), the disadvantage of using carbaryl is that, like DDT, frequent applications increase mite infestations. Removal and burning of infested plants including wild hosts is also recommended. Trellising or stacking structures should be sterilised if they have been used on tomatoes before. Use of sprinkler irrigation dislodges the pests and affords some measure of control, but this has to be balanced against the possibility of increasing the incidence of blight diseases. Biological control of the spider mites by use of predators is at its infancy in Zimbabwe although today a lot more information on predatory mites is available.

## Table 2.1 Acaricides recommended for control of red spider mites in tomatoes in Zimbabwe

| Active ingredient | Trade name and formulation | Code | Type | g/ai | Rate/100l or as stated | PHI | Application details/ Remarks |
|---|---|---|---|---|---|---|---|
| Abamectin | Dynamec 1.8EC | R | EC | 18g/l | 65ml/ | 3 | Spray at first sign of infestation and repeat at 7 day intervals if necessary |
| Amitraz | Mitac 20EC | A | EC | 200g/l | 1500ml/ha<br>300ml | 3 | F.C. S. Repeat at 7 day intervals |
| Clofentezine | Apollo 50 SC | G | SC | 500g/l | 400ml/ha | 4 | Spray at first sign of infestation and repeat as necessary. Mainly an ovicide and larvicide |
| Diazinon | Diazinon 30 EC | R | EC | 300g/l | 190ml | 14 | F.C. S. Repeat at 10 day intervals |
| Dicofol | Kelthane 18.5 EC<br>Dicofol 18.5 EC | R | EC | 185g/l | 220ml | 7 | F.C.S. Repeat as necessary |
| Dimethoate | Dimethoate 40EC | R | EC | 400g/l | 75ml | 14 | F.C.S. Repeat as necessary |
| Malathion | Malathion 1 Dust<br>Malathion 5 Dust<br><br>Malathion 25 WP<br><br>Malathion 50 EC | G<br>G<br><br>G<br><br>G | Dust<br><br>WP<br><br>EC | 10g/kg<br>50g/kg<br><br>250g/kg<br><br>525g/l | R.F.U.<br><br>200g<br><br>120ml | 3<br>7<br><br>7<br><br>7 | Dust as necessary at 100 - 175g/10m$^2$<br>Dust as necessary at 20 - 25g/10m$^2$<br><br>F.C.S. Repeat as necessary<br><br>As above. |
| Sulphur#/Copper | Agridust | A | Dust | 655/65/50g/kg | R.F.U | 14 | Dust plants thoroughly. Repeat at 7 - 10 day intervals |
| Oxychloride#/Malathion# | Vegidust | A | Dust | 655/65/50g/kg | R.F.U | 14 | Dust plants thoroughly. Repeat at 7 - 10 day intervals |
| Oxydemeton-Methyl | Metasystox®25 EC | R | EC | 250g/l | 100ml | 21 | F.C.S. Repeat as necessary. |
| Sulphur | Dusting sulphur | G | Dust | 980g/l | R.F.U | | Dust freely. Repeat as necessary. |
| Propargite | Omite 30 WP<br><br>Omite EC | G<br><br>R | WP<br><br>EC | 300g/kg<br><br>590g/l | 175g<br><br>110ml | 4<br><br>4 | F.C.S. ensuring good under-leaf coverage. |
| Thiometon | Ekatin 25 EC | R | EC | 250g/l | 350-500 | 7 | F.C.S. Repeat as necessary. |

**Key to code:** A = Amber; G = Green; R = Red

**Notes**: PHI= Pre-harvest intervals (days); R.F.U.= Ready for use; F.C.S= Full cover spray; #= Multiple active ingredients in a product

Adapted from Anonymous (2001)

## 2.3. Resistance

Worldwide, cases of resistance have been reported. Resistance is the ability of a population to survive exposure to a crop protection tactic that has been fatal to this population in earlier generations (Dirkse and Van de Vrie, 1996). It is caused by an intensive mortality pressure on a population resulting in selection for individual strains capable of overcoming the burden. Resistance is widely distributed in nature, but occurs at very low frequencies. Since the susceptible strain outnumber the resistant ones, their effects and even their appearance in an area may go unnoticed for years, until an outside pressure, such as the repeated use of a pesticide, eliminates the susceptible biotype (Matthews, 1984). This gives the resistant strain a chance to multiply in the absence of intra-specific competition from the susceptible species. Resistance is therefore different from tolerance in that tolerance acts at species level while resistance can be at a population level, is inherited from parents and never acquired through habituation during the lifetime of an individual pest (Pedigo, 1996) and so can be area specific

According to Pedigo (1996), most documented cases state that resistance originates with mutations occurring regularly in populations and resulting in new genotypes, which are predisposed to resist adverse factors. If the character required for resistance can be obtained through expression of one gene (*monogenic*), resistance may occur after only a few generations, but if many genes, (*polygenic*) are required development is slower. Although all insecticide groups are now affected by this phenomenon, the extent varies greatly between pest species. In some insects, resistance only extends to a few closely-related compounds in a single group; it may be very weak or restricted to a small part of

their geographical range (Pedigo, 1996; Matthews, 1994). At the other extreme, some pests such as the housefly (*Musca domestica*), diamond back moth (*Plutella xylostella*) and Colorado beetle (*Leptinotarsa decemlineata*) now resist most or all of the insecticides available for their control. The most widely used chemical groups, (organochlorines, organophosphates, carbamates and pyrethroids) are those most seriously affected by resistance (Pedigo, 1996). Cross-resistance is when a pest develops resistance to more than one pesticide with similar modes of action, while multiple resistance, is when more than one mechanism is evolved in a pest in response to selection from different insecticide applications. Resistance cannot be produced within a single generation and must be expected against any pest management tactic that imposes a significant burden on a population. Once resistance is fully expressed, continued application of a pest management tactic has no economical benefit on the population (Pedigo, 1996).

### 2.3.1 Conditions that promote resistance

In order to manage resistance it is important to know the factors that influence its development. These were categorised by Taylor and Georghiou (1979) into genetic, biological and operational factors and are summarized in Table 2.2. Not all factors in the table are required at once to produce resistance. However the greater the number present in a situation the greater the chance of occurrence and the more rapidly resistance will develop (Pedigo,1996). Of these factors, genetic and biological factors cannot be changed. Only the operational factors can be modified during IRM. Most of the factors are typical of tobacco spider mites. All genetic and biological factors except that the

species is not highly mobile are specific to tobacco spider mites showing how fast resistance can develop if not managed properly.

**Table 2.2 Factors influencing rate of resistance development**

| | | |
|---|---|---|
| **Genetic factors** | | ➢ Frequency of R alleles<br>➢ Number of R alleles<br>➢ Dominance of R alleles<br>➢ Penetrance, expressivity, interactions of R alleles<br>➢ Past selection by other chemicals<br>➢ Extent of integration of R genome with fitness factor |
| **Biological Factors** | a) Biotic | ➢ The species has a relatively short generation time<br>➢ Monogamy/polygamy, parthenogenesis<br>➢ Numerous offspring per generation are produced |
| | b) Behavioural | ➢ No or little migration occurs between populations<br>➢ The species has a monophagous habit<br>➢ The species is highly mobile increasing the probability of exposure<br>➢ Fortuitous survival, refugia |
| **Operational factors** | a) Chemical | ➢ Insect has a prolonged exposure to a single insecticide or the chemical used is a slow release form<br>➢ The chemical is closely related to the one used earlier<br>➢ The insecticide is inherently irritating and/or repellent |
| | b) Application | ➢ A low population threshold is recommended for application of the insecticide<br>➢ Every generation of the insect is selected<br>➢ Mortality is high (high selection pressure)<br>➢ No functional refugia exist; coverage by the insecticide is effectively complete so that no part of the population remains unselected<br>➢ Large geographical area is covered, all populations in a given area are likely to have been treated<br>➢ Selection occurs prior to mating |

Adapted from: Taylor and Georghiou 1979.

Besides, spider mites possess a remarkable ability to become resistant to most of the pesticide groups in use today (Taylor, 1981). They develop resistance rapidly, particularly where particular pesticides are used for several consecutive seasons (Varela and Seif, 2000). Resistance is difficult to predict but in general, the greater the burden on the population, the faster the rate at which resistance will develop.

### 2.3.2 Mechanisms of resistance

Throughout evolution, insects have had to deal with naturally occurring environmental toxicants. The pre-adaptation hypothesis states that herbivorous pests (like tobacco spider mites) have to detoxify plant defense chemicals and are therefore pre-adapted to detoxify pesticides unlike natural enemies (Grout *et al*, 2000). As stated by Wilkinson (1983) in Pedigo (1996) "insects are forewarned and forearmed to meet the challenges presented them" even by modern synthetic pesticides. To survive they have evolved a variety of mechanisms to make the harmful compounds non-toxic (Pedigo, 1996). There are at least four basic ways that pests overcome the toxicity of pesticides according to Pedigo (1996) and Dirkse and Van de Vrie (1996).

#### 2.3.2.1 Behavioural Resistance

Behavioural resistance is when the insect changes its behaviour in the presence of a pesticide. It is however not very common. According to Pedigo (1996), some are controversial as they often involve metabolic or target site resistance. The question is whether they represent heritable shifts in behaviour or simply survival for a long enough period to exhibit avoidance behaviours. The tobacco bud - worm has acquired this type of

resistance. The resistant larvae slow their movements in the presence of pyrethroids receiving less exposure to lethal doses (Dent, 1991). Mites have also been reported to avoid areas sprayed with pyrethroids (Pedigo, 1996, Dirkse and Van de Vrie, 1996).

#### 2.3.2.2 Physiological Resistance

Physiological resistance is any form of resistance that reduces toxicity through changes in the basic physiology of the pest that includes a decrease in penetration of the pesticide through the body wall or increases in the rate of excretion of the chemical (Pedigo, 1996).

#### 2.3.2.3 Biochemical/ Metabolic Resistance

The pesticide is attacked by one or more enzymes that detoxifies it before it reaches its site of action resulting in insects excreting the pesticide as a primary or secondary product. According to Pedigo (1996), biochemical resistance is most common in insects.

#### 2.3.2.4 Target site insensitivity

Pests may also develop a change in the target site eliminating the intended effect of the pesticide. Target site insensitivity is when the pesticide is not metabolised more rapidly even if it penetrates the insect cuticle and does not kill the pest, so the target site is insensitive. Altered acetylcholinesterase makes the cattle tick, mosquito (*Aedes albimanus*) and the two-spotted spider mite (*T. urticae*) resistant to certain organophosphates. The pest may lack a metabolic site of action. Thus, the pesticide fails to kill the pest

### 2.3.3 Strategies for managing and slowing/delaying resistance

Resistance management aims at maintaining the usefulness of insecticides and begins with recognizing the factors that influence its development. It must therefore begin before detection efforts confirm that resistance development is underway. There is need to minimize selection pressure to keep susceptible insects alive so that the genes for susceptibility are a valuable natural resource maintained. Growers must realize that once heavy infestations occur good control is economically impossible. As stated by Pedigo (1996) prevention of resistance to any effective pest management tactic may be practically impossible in many situations. Resistance management must therefore be a matter of anticipating and slowing the rate of development. The rate of resistance development can however be slowed down by considering the operational factors that enhance it, and then modify the pest management program accordingly.

The most basic routine is to combine control tactics to achieve suppression (chemical, biological, cultural, mechanical). Proper IPM methods with the help of biological control agents such as parasites and predators need to be considered (Dirkse and Van de Vrie, 1996). Decreasing pesticide use through IPM is more productive than any other IRM strategy therefore optimize resistance management through promoting IPM. Efficient scouting for initial infestation to enable early detection of mites is of prime importance. Multiple tactics place diverse pressures on the pest population making it more difficult for the species to overcome the effects of any one tactic. If resistance still develops to one of the tactics in the integrated scheme, its effects will be lessened because other tactics still contribute to suppression. Good crop management practices that include proper

irrigation, fertilization and general good crop husbandry produce vigorous plants that tolerate insect injury better.

## 2.4 Natural enemies of vegetable pests

Pests rarely reach damaging levels under natural conditions as natural enemies keep them under control. Several predacious mites from the families Stigmaeidae, Phytoseidae, and Tydeidae and some insects are known to feed on phytophagous mites. The staphylinid beetles (*Oligota spp*) are known to feed on this species (Varela and Seif, 2000). Three species of beetles belonging to the genus *Stethorus* are important predators of *T. urticae* in orchards in Australia (Dent, 1991). Studies in unmanaged orchards have shown that the European red mite (*Panonychus ulmi*) and *T. urticae* mite populations are strongly regulated by predatory mites and other natural enemies (Pedigo, 1996; Varela and Seif, 2000). Successful natural enemies against *T. urticae* in glasshouse crops in Europe and America are predatory mites, *Phytoseiulus persimilis* and *Amblyseius anderson* (Dent, 1991; Varela and Seif, 2000). The predatory mite (*P. persimilis*) is one of the most voracious predatory species. Each adult may eat 5 adult phytophagous mites or 20 nymphs plus several eggs per day. The predator *Amblyseius fallacis/ andersoni* (Chant) can virtually eliminate phytophagous mites from apple trees according to Pedigo (1996). Other mite predators include *Chrysopa* larvae (lacewing larvae), predaceous thrips, predaceous staphylinid beetles (Coleopteran which unfortunately prefers the cooler months) and Coccinellidae (ladybird beetles), anthocorid bugs, mirid bugs and cecidomyid and syrphid flies (Brettell and Burgess, 1981; Varela and Seif, 2000; Magdalena & Meyer 1996). According to Varela and Seif (2000), several species of

predatory mites are known from Eastern and Southern Africa but there are no detailed investigations in their role in tomato fields yet. The common natural enemies of vegetable pests in Southern Africa as listed by Verkerk (2001) are given in Table 2.3.

**Table 2.3 Major natural enemies of common vegetable pests.**

| Name | Main prey | Crops |
|---|---|---|
| | ***Predators*** | |
| Ladybird beetle: (Coleoptera-Coccinelidae) | Aphids, mites, thrips, small caterpillar, insect eggs | Brassicas, tomatoes, potatoes, beans |
| Ground beetles (Coleoptera-Carabidae) | Grubs, eggs | Most crops |
| Hoverfly:(Diptera –Syrphidae) | Aphids, small caterpillars, eggs | Most vegetables |
| Predatory wasps- (Hymenoptera- | Caterpillars and sawfly larvae | Most crops |
| Praying mantis- Orthoptera-Mantodae) | Flies, small caterpillars, aphids | All crops |
| Predacious ants (Hymenoptera-Formicidae) | Caterpillars, eggs, pupae of arthropods | Most crops |
| Dragon flies/Damselflies - (Odonata- ) | Moths, caterpillars, nymphs, aphids, mosquitoes | Most crops |
| Chameleon, frogs, snakes | Crickets, aphids, caterpillars, locusts | Most crops |
| Rove beetle: (Coleoptera - Staphylinidae) | Insect eggs, mites, aphids, scale insects, small insects. | All crops and on soil |
| Pirate bugs: (Hemiptera - Anthoridae) | Thrips, mites, aphids, Insect eggs, small caterpillars | Beans, potatoes, tomatoes |
| Predatory mites: (Acarina-Phytoseiidae) | Phytophagous mites, thrips, insect eggs | Tomatoes, eggplants, beans |
| Lacewing larvae/(Aphid lion-Chrysopadae) | Aphids. Mites, small caterpillars & pests | Beans, tomatoes, potatoes. |
| Spiders: Arachnida (Araneae) | Flies, moths, small caterpillars, mites, aphids. | Most crops |
| Entomopathogenic Nematodes (Steinermema spp) | Spodoptera caterpillars, nematodes, grubs, cutworms | All crops |
| | ***Parasitoids*** | |
| Parasitoid wasp: Hymenoptera (many families) | All pest insects, caterpillars, aphids, bugs, eggs, pupae | All crops |
| Whitefly parasite (*Encarsia formosa)* | Whitefly | Tomato, potato |
| | ***Parasites*** | |
| Fungus | Diamond back moth larvae, caterpillars etc | Brassicas, tomatoes |
| Trichoderma spp | Soil borne insects, Stem borers | Solanaceous crops |
| Viruses (Granulosis virus) | Diamond back moth larvae, caterpillars | Brassicas |
| Nuclear Polyhedrosis Virus (NPV) | Semi-looper, caterpillars | Brassicas, tomatoes |

Adapted from: Verkerk 2001

### 2.4.1 Effects of pesticides on natural enemies

Field observations have shown that natural enemies are capable of controlling infestations provided they are not disturbed by the severe use of broad-spectrum insecticides and the crop is properly irrigated (Pedigo, 1996; Varela and Seif, 2000). The indiscriminate use of broad-spectrum insecticides eliminates these natural enemies (Varela and Seif, 2000). Most insecticides used today are broad-spectrum in action hence are toxic to the beneficial ones. Elimination of predators stimulates mite outbreaks (Duncombe, 1976; Pedigo, 1996; Varela and Seif, 2000). The food limitation hypothesis, states that pesticides kill the prey decreasing the amount of food for natural enemies, which then die or emigrate, leaving resistant prey strains an abundant food source (Grout *et al.* 2000, Verkerk 2001). Very often, the phytophagous mites have developed resistance to many of the broad-spectrum insecticides used, but predators usually succumb to the pesticide (Pedigo, 1996; Verkerk, 2001). This is probably because of the pre-adaptation hypothesis tobacco spider mites detoxify pesticides, unlike natural enemies (Grout *et al.* 2000).

#### 2.4.1.1 Lethal effects

Many natural enemies are more mobile than their prey or host therefore come into physical contact with more pesticide residues. They consume numerous prey items of which each may contain sub-lethal concentrations of pesticides hence exposed to far higher pesticide levels than individual prey items. High mortality of natural enemies occurs from direct exposure to the pesticide. Natural enemies are more sensitive to pesticides than pests because most are smaller than their prey, hence take up more

pesticides in proportion to their body volume. Most natural enemies move around the sprayed surfaces more than their hosts which are generally stationary when feeding, hence pick up more chemical (Verkerk, 2001). Besides they cannot detoxify poisons very well as pests are better adapted for detoxification since they already possess the enzymes necessary for breaking down natural poisons found in some of the plants they eat. On the other hand, carnivorous insects do not have the same levels of such enzymes. Due to their susceptibility to pesticides, it is expected therefore that spraying broad-spectrum compounds harm natural enemies more than pests. Pesticides therefore reduce natural enemies directly and indirectly. To avoid killing some natural enemies specific acaricides such as dicofol, tetradifon and binapacryl have often been recommended in cotton according to Taylor (1981). Different acaricides therefore have different effects on different natural enemies (Table 2.4)

#### 2.4.1.2 Sublethal effects

Apart from direct effects of mortality, pesticides can be harmful by interfering with fecundity or searching behaviour. It is better therefore to use selective acaricides and avoid broad-spectrum pesticides. Natural enemies may however adapt and become behaviourally or biochemically tolerant to pesticides. Croft (1979) in Matthews (1984) pointed out that where organophosphates have been used in apple orchards for many years, growers have actually benefited from the mite predator *Typhlodromus occidentalis* becoming tolerant to the compounds in the North Western United States of America (Matthews, 1984).

**Table 2.4 Known effects of some pesticides on some natural enemies**

| Compound | *Phytoseiulus persimilis* | | *Encarsia formosa* | | *Typhlodromus spp* |
|---|---|---|---|---|---|
| | Eggs | Adults | Pupae | Adults | Adults |
| Clofentezine | | S | | S | S |
| Cyhexatin | | H | S | S | S |
| Deltamethrin (Decis) | H | H | H | H | H |
| Demeton-S-methyl | H | H | S | H | S |
| Diazinon | S | I- H | H | H | |
| Dichlorvos | | H | | H | |
| Dicofol | | H | S | I | H |
| Dicofol/ tetradifon | I | | I | I | |
| **Dimethoate** | H | H | H | H | H |
| Endosulfan | | H | | | |
| Gamma-HCH (lindane) | H | H | H | I | S |
| Heptenophos | | H | S | I | |
| **Malathion** | H | H | H | H | |
| Oxydemeton-methyl | | H | | | |
| Parathion | H | H | H | H | |
| Permethrin (Ambush) | | H | S | H | H |
| Petroleum emulsion | S | S | S | I | |
| Propargite (Omite) | | | | | S |
| Pirimicarb | S | | I | | S |
| Pyrethrum/ resmethrin | H | H | | H | |
| Tetradifon | S | S | S | S | S |
| **Abamectin** | | H | | H | H |
| **Amitraz** | | H | | H | H |

Adapted from: Matthews, 1984; Verkerk, 2001.
Key: H = Harmful I = Intermediate S = Safe

### 2.4.2 Consequences of eliminating natural enemies

In addition to resistance, pest resurgence and replacement are other phenomena associated with the use of conventional pesticides in a number of agricultural systems. However, the possibility of their occurrence with any pest management tactics is there particularly if the tactic is directly favourable to the physiology of the insect pest or has an adverse effect on important natural enemies. There are three possible causes of pest resurgence and replacement namely: reduction of natural enemies by pesticides along with the pest direct favourable influences of pesticides on physiology and behaviour of arthropods (hormoligosis) and the removal of competitive species. Of these, upsets from

the reduction of natural enemies after insecticide application presents most experimental evidence.

### 2.4.2.1. Resurgence

This can be defined as a situation where a population, after having been suppressed, re-bounces to numbers greater than before suppression occurred. The broad spectrum activity by insecticides against insect species even at low dosages presents a risk to beneficial and other non-target insects causing resurgence of the pest species or pest outbreak of another species (Matthews, 1984). After spraying a broad-spectrum insecticide, pests, natural enemies and other non-target insects are killed. The few pests that escape the effects of the chemical whether by chance or due to resistance will increase in numbers faster without any intra-specific and inter-specific competition.

### 2.4.2.2. Replacement or secondary pest outbreak

This occurs when a major pest is suppressed and continues to be suppressed by a tactic but is replaced by another pest previously with minor status. In Zimbabwe use of DDT and carbaryl in cotton to control the African and red bollworms, white fly, jassids and lygus as the major pests resulted in an increase in red spider mites which used to be a minor pest then.

# CHAPTER THREE

# MATERIALS AND METHODS

## 3.1. Laboratory Rearing Techniques

### 3.1.1. Test mites

Tobacco spider mites (*Tetranychus evansi*)

Adults of the suspected resistant strains were collected from two locations Chinamhora and Mutoko. Potted tomato plants were infested with these mites for multiplication purposes. The susceptible strain was obtained from the Agricultural Research Centre Harare. This population had not been sprayed for over two years. The second-generation adults of the susceptible population were used to conduct bioassays, with each of the four acaricides, abamectin (Dynamec 1.8 EC), amitraz (Mitac 20 EC), malathion (Malathion 50 EC) and dimethoate (Dimethoate 40 EC) (Table 3.1). The suspected resistant strains were then exposed to three doses of each acaricide.

### 3.1.2. Natural enemies

Predatory mites, *(Phytoseiulus persimilis)*

Young heavily infested leaflets from the prey culture were removed, cutting them off at the base of the stalk. They were placed upper surfaces facing upwards on pads of wet cotton wool in petri dishes. The leaf stalks were stuck into cotton wool to keep the leaves fresh. Wool was pulled up with forceps and pressed against the leaf edges to avoid mites getting underneath the leaves. Predatory mites were collected from Fambidzanayi Permaculture Centre and added to each infested leaf using a moistened brush aiming for a

ratio of 2: 1 prey: predator. The petri dishes were placed in a tray part filled with water to prevent mites from escaping and left in the laboratory at a temperature of about 28° C and 60% humidity. The cotton wool remained wet to sustain the leaf. Predator eggs were transferred each day to new rearing petri dishes to give cultures of young mites of roughly the same age and help decrease cannibalism. The adult females were then transferred to new dishes to lay eggs and prevent overlapping of generations in the same dish. Newly hatched predatory mites were fed from the prey culture and used in the bioassays.

Ladybird beetle *(Hippodamia variegata)*

A rape crop was raised in the green house at the Agricultural Research Centre in Harare, and infested with aphids. Ladybird beetle adult males and females were collected from the fields in Chinamhora. One male and two females were placed into empty peanut butter plastic containers measuring 6.5 cm diameter and 12.5 cm in height and lined with filter paper with holes on lids as mating containers. Rape leaves with aphids were placed in the containers as a source of food for the beetles. Leaves were replaced often. On alternate days, beetles were provided with cotton wool soaked in a honey solution as an extra source of fluid and energy important for females during the pre-oviposition period. Eggs were removed and placed on moistened filter paper in small petri dishes with holed lids. Aphids were added to feed larvae as they hatched. Pupating larvae were kept in empty plastic containers until adults emerged. Emerging adults were also fed with aphids. Their outer wings were left to harden and darken and then used in the bioassays.

Aphid wasp parasitoid, *(Diaeretiella rapae)*

Rape leaves with parasitised aphids were collected from fields in Chinamhora and put in cardboard boxes, which acted as the incubation chamber for the parasitoids to emerge (Plate 3.1). Two holes large enough to allow clear 500 ml empty plastic soft drink bottles to be fitted in were cut out. A sugar solution was used to wash the bottles so that the adult parasitoids could feed from the dried sugar solution. Light into the cage was only through the plastic bottles. On emerging from the aphids, the adult parasitoids were trapped in the bottles as they flew towards light. Once the bottles were 'filled' with adult parasitoids they were removed from the chambers and their tops quickly covered and replaced with new empty bottles. These adults identified as *Diaeretiella rapae* by a taxonomist at the National Museum in Harare were used in the bioassays.


**Plate 3.1** Incubation chamber (*Courtesy of Musundire 2002*)

## 3.2 Laboratory Bioassay Procedure

The objectives of the bioassays were

- To detect and measure resistance of the tobacco spider mites to currently recommended and mostly used acaricides
- To determine the effects of these acaricides on common vegetable natural enemies

### 3.2.1 Tobacco Spider Mite

*Leaf-disk technique*

The experiment was conducted in the laboratory at the Agricultural Research Centre in Harare. The acaricides used in the bioassays and their modes of action are given in Table 3.1 while a range of the six concentrations tested are given in Table 3.2.

A leaf disk cutter was used to cut leaf disks of 2 cm diameter from mite free tomatoes raised separately in a glasshouse. One hundred and thirty-five (135) cosmetic pads were soaked in distilled water and each put in a petri dish. Each concentration was replicated five times. The leaf disks were then dipped in respective concentrations for 3 seconds and dried for about an hour. The disks were each placed on the wet cosmetic pads with their undersides facing upwards. Ten (10) susceptible female mites were placed on each leaf disk using a small soft brush. The petri dishes were kept in the humid chamber at about 25°C and about 50% RH with constant illumination. Mortalities were recorded after 24. 48, 72 and 96 hours. The lethal concentration ($LC_{50}$) at which 50% of the susceptible mites died was selected for each acaricide.

**Table 3.1: Acaricides used**

| Common name | Trade name | Chemical group | Mode of action against arthropods |
|---|---|---|---|
| Abamectin | Dynamec 1.8EC | Avermectin | ▪ Contact and stomach action with limited plant systemic activity but exhibits translaminar movement<br>▪ Acts by stimulating the release of γ-aminobutyric acid an inhibitory neurotransmitter causing paralysis in motile stages of mites, leafminers and Colorado beetles. |
| Malathion | Malathion 50EC | Organophosphate | ▪ Non-systemic with contact, stomach and respiratory action<br>▪ Cholinesterase inhibitor against the Acari, Coleoptera, Diptera, Hemiptera, Hymenoptera and Lepidoptera. |
| Amitraz | Mitac 20EC | Amidine | ▪ Non- systemic with contact and respiratory action.<br>▪ Interferes with octopamine receptors in the tick nervous system causing an increase in nervous activity against *Tetranychid* and *Eriophyid* mites, pear suckers, scale insects, mealy bugs, whiteflies, aphids, eggs and larvae of Lepidoptera. |
| Dimethoate | Dimethoate 40EC | Organophosphate | ▪ Systemic insecticide and acaricide with contact and stomach poison.<br>▪ Cholinesterase inhibitor against Acari, Aphididae, Aleyrodidae, Coccidae, Coleoptera, Collembola, Diptera, Lepidoptera, Pseudococcidae and Thysanoptera |

**Table 3.2 Concentrations of the various formulations used in the leaf disk technique**

| Treatments | Concentration (gai/l) | | | |
|---|---|---|---|---|
| | **Abamectin** | **Malathion** | **Mitac** | **Dimethoate** |
| T1 | 0.0000000 | 0.0000000 | 0.0000000 | 0.0000000 |
| T2 | 0.0014625 | 0.0790000 | 0.0750000 | 0.0380000 |
| T3 | 0.0092500 | 0.1575000 | 0.1500000 | 0.0750000 |
| T4 | 0.0058500 | 0.3150000 | 0.3000000 | 0.1500000 |
| T5 | 0.1170000 | 0.6300000 | 0.6000000 | 0.3000000 |
| T6 | 0.0234000 | 1.2600000 | 1.2000000 | 0.6000000 |
| T7 | 0.0351000 | 1.8900000 | 1.8000000 | 0.9000000 |

Calculation for active ingredients in spray mixture $= \frac{\text{Formulation rate/l}}{1000\ \text{ml}} \times \text{gai/l}$

*The leaf-dipping technique*

Thirty tomato plants about 15 - 30 cm high were selected from a stock free of spider mites and placed in the laboratory. Unwanted leaves were removed leaving five healthy ones on each plant. Of the 30 plants, 15 were labeled for the Chinamhora population, the other 15 for Mutoko. Each plant was then labeled for a different acaricide and concentration. Four concentrations for each acaricide including the $LC_{50}$ were prepared as given in Table 3.3. The five leaves were dipped singly for 3 seconds into the appropriate treatment and distilled water and left to dry. Ten female tobacco spider mites from each relevant location were placed on each leaf. The five (5) leaves per plant were treated as replicates. Petroleum jelly was ringed around the leaf petioles to deter the mites from wandering off the treated leaves. The tomato plants were left in the laboratory with temperatures set at about 25°C and 50% RH. Constant illumination during this period also limited mite dispersal. Mortality assessments were carried out after 96 hours. The mites were transferred onto a sheet of white paper on which the number of dead mites was easily determined with the aid of a binocular microscope.

**Table 3.3 Concentrations of the various formulations used in the leaf dip technique**

| Treatment | Concentration gai/l | | | |
|---|---|---|---|---|
| | Abamectin | Malathion | Mitac | Dimethoate |
| T1 | 0.00000 | 0.00000 | 0.00000 | 0.00000 |
| T2 | 0.00585 | 0.63000 | 0.30000 | 0.30000 |
| T3 | 0.01170 | 1.26000 | 0.60000 | 0.60000 |
| T4 | 0.02340 | 1.89000 | 1.20000 | 0.90000 |

### 3.2.2 Natural enemies of vegetable pests

The field recommended acaricides rates were used for all the natural enemy bioassays (Table 3.4).

**Table 3.4 Recommended field rates for selected acaricides against mites on tomato**

| Rate | Acaricide | | | |
|---|---|---|---|---|
| | Abamectin | Malathion | Mitac | Dimethoate |
| ml/100l | 65.0 | 120 | 300 | 75.0 |
| gai/l | 18.0 | 525 | 200 | 400 |

Predatory mites, *(Phytoseiulus persimilis)* (According to Hussey and Scope 1985)

Damp cosmetic pads were each placed in 15 petri dishes for each of the 4 acaricides. A piece of double-sided sticky tape was stuck onto 15 slides close to the ends to make a sticky base for attaching predatory mites. Three (3) slides for each treatment were made. Using a wetted brush 20 predatory mites were placed on their backs on the sticky base of each slide arranging them in 4 rows of 5 mites each. The mites' legs were left free from the sticky tape as leg movement indicated survival. Slides were dipped into the prepared acaricide solutions for 3 seconds so that the predatory mites were completely immersed in the mixture. The dipped slides were drained to remove remaining droplets and placed on a drying rack made from folding pieces of cardboard paper. Slides were placed on racks in the humid chamber. Dipped mites were examined using a stereoscopic

microscope after 24 hours. The dead mites were recorded on each slide. They were touched with a small brush and if they did not move their legs or mouthparts, they were recorded as dead. The mortality for each acaricide was calculated

Ladybird beetle (*Hippodamia variegata*)

Ten adult beetles were placed in each of the 20 plastic peanut butter jars measuring 6.5 cm in diameter and 12.5 cm in height. Rape leaves infested with aphids were sprayed with 50 ml of prepared acaricide using a 500 ml hand sprayer and left to dry for about an hour. Some of these were sprayed with distilled water as the control. The dry treated leaves were then put in respective containers with the adult beetles. The containers were closed, placing a piece of tissue paper between the container and the holed lid to prevent condensation forming inside. These were left in the lab at room temperature. Conditions of the predators were checked and recorded after 48 hours with a count of dead insects. After 48 hours, the insects were gently touched with a brush to determine motion. If they walked off in an abnormal manner, they were counted as dead. The percentage of the beetles killed by each acaricide was recorded.

Aphid wasp parasitoid (*Diaeretiella rapae*):

The respective recommended acaricide concentrations were prepared and small amounts put in labeled clear soft drink bottles (exposure bottles) and shaken to make sure the inner surfaces were treated. Excess acaricides were poured out and the treated exposure jars left open to dry. The bottles (emergence bottles) from the incubation chamber 'filled' with aphid wasp parasitoids were removed and the lids quickly replaced. They were wrapped

with a black cloth to limit movement of the parasitoids. The lid of an empty exposure bottle was removed and the exposure bottle quickly put over the neck of the emergence bottle with parasitoids making sure there was no gap through which the parasitoids could escape. The treated exposure bottles were exposed to light and 10 parasitoids let in upwards into the treated bottles. The bottles were quickly closed tightly. When the parasitoids were settled down, a wad of cotton wool soaked in 10% sugar solution was introduced into the exposure bottles as food source for the adults. After 48 hours, the exposure bottles were covered with black cloth. The lid was quickly removed placing an empty collecting bottle over the mouth of the exposure bottle and light shown downwards onto the collecting bottle. The live parasitoids flew up into the collecting jar and the number of the dead parasitoids recorded.

### 3.2.3 Evaluation of bioassay results

*3.2.3.1 Resistance status of tobacco spider mite*

The procedure for computing the regression line and $LC_{50}$ for all acaricides was done using the probit analysis according to Finney (1962). Data on the relationships between doses and mortalities were obtained and graphs and mathematical calculations used to estimate the parameters in the probit analyses. These graphs (Figures 4.1 to 4.4) and the mathematic calculations depended upon the probit transformations. In order to make the estimate, the percentage kill observed for each concentration for each acaricide were calculated and converted to probits. These were plotted on a graph paper and lines of best fit by the eye drawn. The lines were then used to initiate the mathematical calculations for better fitting lines. The slopes (b-values) of the lines were estimates of the inverse of

the standard deviations obtained as the increase in Y for the unit increase in x. The equation: $y = Y + b(x- X)$ where y = expected probit; Y= mean of product of the weight of the working probit; x = log dose (+a) and X= mean of product of weight of x, was used. The chi-square tests according to Fisher (1944) were used to test representations of data by the lines using the following formula: $X^2 = \sum (O-E)^2 / E$ where O = Observed and E = expected. The $LC_{50}$ for each of the mite populations were calculated by determining the value of x when y = 5. The actual percentage lethal concentration was calculated by finding the antilog of the value of x. Further calculations were done for the chi-squares, which gave an indication of whether the experimental points determined a straight line, in order to see if expected results fitted the observed results. Variance and Fiducial limits were calculated to evaluate the accuracy of the bioassays. In the assessment of the resistance status of *T. evansi* to the acaricides the $LC_{50}$ values for the susceptible and suspected resistant populations were compared and the Resistant Factors (RF) calculated as follows:

$$RF = \frac{LC_{50} \text{ (suspected resistant population)}}{LC_{50} \text{ (susceptible population)}}$$

Where mortalities occurred in the control treatments corrections were done using Abbott's formula (Abbott 1925) as follows:

$$P = \frac{Po - Pc}{100 - Pc} \times 100$$

where: P = Corrected mortality

Po = Observed mortality

Pc = Mortality of control treatment

*3.2.3.2 Effects of acaricides on natural enemies of vegetable pests*

The percentage mortalities were worked out for each treatment. Based on these results each acaricide was placed into one of the four pesticide risk categories for natural enemies (Table 3.5).

**Table 3.5 Pesticide risk categories for natural enemies**

| Risk category | | Corrected % killed |
|---|---|---|
| | Harmless | < 30 |
| | Slightly harmful | 31-79 |
| | Moderately harmful | 80-99 |
| | Harmful | >99 |

Adapted from Anonymous (2000)

## 3.3 Acaricide Field Trials

The objectives of the field trial were:

- To confirm the laboratory findings on the resistance status of the tobacco spider mite to selected acaricides under field conditions
- To confirm laboratory findings on the effects of the same acaricides on vegetable natural enemies under field conditions

### 3.3.1 Sites

The study was carried out in Chinamhora at two sites, Munyawiri and Govera wards. Two farmers' fields were used. The crop was planted during the first week of July 2002.

### 3.3.2 Treatments

The four acaricides used in the bioassays were used in the field. The acaricides were applied weekly using the recommended rates of 65 ml/ha abamectin, 1500 ml/ha amitraz, 75 ml/ha dimethoate and 120 ml/ha malathion.

### 3.3.3 Overall treatments

The tomato cultivar Rodade was used. Seedlings were raised in the greenhouse at the Agricultural Research Center, Harare. Twenty plots each measuring 5m x 4m were prepared and tomatoes planted in rows 90 cm apart and 45 cm in-row spacing. Compound S (7N: 21P: 7K) fertilizer at a rate of 1 000 kg/ha was applied as basal. Ammonium nitrate (34.5N) was then applied at 90kg/ha rate in two split applications, the first half at establishment and the second at early fruit bearing stage. At planting, confidor (200 g a.i./l) at a rate of 500 ml/ha was drenched to control leaf eating and other chewing pests. Dithane M45 was used as preventative measure to control blights and leaf-eating pests were later controlled using carbaryl 85 WP. No artificial infestation of mites was done, as Chinamhora is an area with naturally high mite pressure. First acaricide sprays were applied as soon as mites were noticed and every other week thereafter. At the end of the trial each acaricide had been applied 5 times.

### 3.3.4 Trial design

The design was a complete randomised block design (CRBD) with treatments replicated 5 times. Each plot had five rows with about 50 plants. A pathway measuring 1.5m was left around plots.

### 3.3.5 Parameters measured

Data was collected from the 3 middle rows excluding the outer rows and the first and last two plants in each of the three middle rows. Weekly samplings were done for six weeks from establishment. Two plants per row were selected randomly. Each selected plant was divided into three sections i.e. top, middle and bottom. From each section, two fully open leaflets were removed from the plant. The leaflets were taken to the laboratory in individual khaki envelopes where mites were counted. Damage levels caused by spider mites were determined by using a damage leaf index ranking from 1-5 where 1 represented a few spots and 5 for a leaf totally covered with spots with dry patches occurring after Hussey and Scopes (1985). The parameters included tobacco spider mite weekly populations, the different natural enemies and other insects in the plots. The effects of the acaricides on these insects in the field were recorded.

### 3.3.6. Evaluation of field data

Total mite counts for each treatment per week were tabulated for each site. ANOVA on treatment means was conducted using the MSTAT programme.

# CHAPTER FOUR

# RESULTS

## 4.1 Laboratory Bioassays

### 4.1.1 Resistance status of tobacco spider mites

The laboratory bioassay results for abamectin, malathion, amitraz and dimethoate against the tobacco spider mite are given in Table 4.1.

**Table 4.1 Bioassay results for selected acaricides against the tobacco spider mite**

| Strain | Statistical parameters | Abamectin | Malathion | Amitraz | Dimethoate |
|---|---|---|---|---|---|
| Susceptible | $LC_{50}$ (gai/l) | 0.0306 | 0.1000 | 0.0300 | 0.0760 |
| | Slope ±SE | 2.5 ± 0.007 | 3.0 ± 0.005 | 1.6 ± 0.009 | 2.5 ± 0.018 |
| | F.L (95%) | 1.60 - 1.80 | 1.90 - 2.07 | 1.40 - 1.60 | 1.70 - 2.20 |
| | $X^2$ | 0.10523 | 0.05301 | 0.00879 | 0.02757 |
| Chinamhora | $LC_{50}$ gai/l | 0.0360 | 0.1000 | 0.0420 | 0.1080 |
| | Slope ±SE | 2.9 ± 0.006 | 2.5 ± 0.008 | 2.5 ± 0.007 | 2.0 ± 0.007 |
| | F.L (95%) | 1.92 - 2.08 | 2.20 - 2.40 | 2.01 - 2.20 | 2.40 - 3.00 |
| | $X^2$ | 0.01906 | 0.00233 | 0.00939 | 0.04086 |
| | RF | 1.2 | 1.0 | 1.4 | 1.4 |
| Mutoko | $LC_{50}$ gai/l | 0.0360 | 0.1200 | 0.0420 | 0.9200 |
| | Slope ±SE | 2.6 ± 0.007 | 2.5 ± 0.013 | 1.9 ± 0.013 | 2.2 ± 0.008 |
| | F.L (95%) | 1.91 - 2.09 | 2.20 - 2.50 | 1.92 - 2.28 | 2.20 - 2.40 |
| | $X^2$ | 0.00410 | 0.01259 | 0.01182 | 0.14145 |
| | RF | 1.2 | 1.2 | 1.4 | 12.1 |

**Key:** $LC_{50}$ denotes the median lethal concentration

Slope = b-value of lines on the probit regression lines

RF= $\frac{LC_{50} \text{ for suspected resistant population}}{LC_{50} \text{ for susceptible population}}$

F.L denotes fiducial limits

Tabular chi-square = 7.815

Based on the $LC_{50}$ values, amitraz ($LC_{50}$ = 0.0300 g a.i./l) and malathion ($LC_{50}$ = 0.1000) were the most and least efficacious acaricides respectively against the susceptible tobacco spider mite strain. In contrast, abamectin and dimethoate were the most and least efficacious respectively against the suspected resistant populations from Chinamhora and Mutoko.

Populations from Chinamhora had RFs ranging from 1.0 (malathion) to 1.4 (amitraz) whilst those from Mutoko ranged from 1.2 (abamectin) to 12.1 (malathion). These results indicated low levels of resistance development by the tobacco spider mite against the these acaricides especially dimethoate.

### 4.1.2. Effects of acaricides on selected natural enemies of vegetable pests

Results of the effects of acaricides on selected natural enemies of vegetable pests are given in Table 4.2.

**Table 4.2 Effects of selected acaricides on natural enemies of vegetable pests**

| Acaricide | Natural enemy (% mortality) | | | |
|---|---|---|---|---|
| | **Ladybird beetle** | **Aphid wasp parasitoid** | **Predatory mite** | **Mean** |
| Abamectin | 95.0 | 98.0 | 100.0 | **96.70** |
| Malathion | 72.5 | 94.0 | 96.0 | **87.50** |
| Mitac | 90.0 | 96.0 | 98.0 | **94.70** |
| Dimethoate | 77.5 | 98.0 | 98.0 | **91.20** |
| **Mean** | **83.80** | **96.50** | **98.00** | **92.80** |

Overall, all the four acaricides were harmful to all the three natural enemies. Mortalities ranged from 72.5 percent (malation on the ladybird beetle) to 100.0 percent (abamectin on the predatory mite). On average, all the acaricides were least harmful on the lady bird

beetle (83.80 percent mortality) and most harmful on the predatory mite (98.00 percent mortality). Also on average, malathion and abamectin were the least (87.50 percent mortality) and most (96.70 percent mortality) harmful respectively on all the natural enemies tested.

The acaride risk categories for the natural enemies tested are shown in Table 4.3.

**Table 4.3 Acaricide risk categories for the natural enemies tested**

| Acaricide | Ladybird beetle | Aphid wasp parasitoid | Predatory mite |
|---|---|---|---|
| Abamectin | | | |
| Malathion | | | |
| Amitraz | | | |
| Dimethoate | | | |

**Key:**

| Risk category | | Corrected % killed |
|---|---|---|
| | Harmless | < 30 |
| | Slightly harmful | 31-79 |
| | Moderately harmful | 80-99 |
| | Harmful | >99 |

None of the acaricides tested against the three natural enemies could be categorized as harmless to any one of them. Abamectin was categorized as moderately harmful to the ladybird beetle and aphid wasp parasitoid and harmful to the predatory mite. Malathion and dimethoate were both categorized as slightly harmful to the ladybird beetle but moderately harmful to the other two natural enemies. Amitraz was categorized as moderately harmful to all the three natural enemies.

## 4.2 Acaricide Field Trial

### 4.2.1 Acaricide efficacy against the tobacco spider mite under field conditions

At the Munyawiri Ward site, data was collected up to week 4 only because of an outbreak of russet mites, which wiped out the whole crop leaving the abamectin treatments only. However, this happened after fruiting and so the other treatments had most leaves twisting and turning bronze coloured and fine cracks appearing on fruits typical of russet mite damage. This resulted in generally lower populations of tobacco spider mites from site one. Scoring for tobacco spider mite damage became impossible as leaves were badly folded, dried and defoliation resulted. The developed fruits were exposed to sunlight due to defoliation resulting in sunscald. At the Govera Ward site, bollworms attacked the crop initially but were controlled using carbaryl (Carbaryl 85 WP). There were no incidences of russet mites at this site. The ANOVA results on the efficacy of the four acaricides on the tobacco spider mite data for the two sites are presented in Tables 4a and b.

**Table 4.4a Efficacy of four acaricides on tobacco spider mite field populations at the Munyawiri Ward site, Chinamhora**

| Treatment | July | August | | September | |
|---|---|---|---|---|---|
| | Week 1 | Week 2 | Week 3 | Week 4 | Means |
| Abamectin | 65 (0) | 109 (0) | 94 (0) | 433(1) | 212.00 **ab** |
| Malathion | 37 (0) | 95 (0) | 439 (2) | 567(3) | 367.00 **b** |
| Amitraz | 48 (0) | 85 (0) | 92 (0) | 248(2) | 141.67 **a** |
| Dimethoate | 71 (0) | 93 (0) | 501(2) | 597(3) | 397.00 **bc** |
| LSD (0.05) | | | | | 238.058 |
| P-value | | | | | 0.1065 |
| CV (%) | | | | | 42.64 |

Notes:
Figures in brackets were average damage scores for the different treatments.
Treatment means followed by different letters are statistically significantly different at $P > 0.05$

**Table 4.4b Efficacy of four acaricides on tobacco spider mite field populations at the Govera Ward site, Chinamhora**

| Treatment | July | August | | September | | October | | Means |
|---|---|---|---|---|---|---|---|---|
| | Week 1 | Week 2 | Week 3 | Week 4 | Week 5 | Week 6 | Week 7 | |
| Abamectin | 78(0) | 111(0) | 94(0) | 440(2) | 119(2) | 89(2) | 477(3) | 216.667**ab** |
| Malathion | 81(0) | 64(0) | 451(2) | 576(3) | 95(2) | 448(2) | 602(3) | 368.667 **b** |
| Amitraz | 50(0) | 90(0) | 92(0) | 271(2) | 88(2) | 114(2) | 295(3) | 154.833**a** |
| Dimethoate | 70(0) | 94(0) | 510(2) | 867(4) | 90(3) | 553(3) | 595(4) | 451.500 **bc** |
| LSD (0.05) | | | | | | | | 155.621 |
| P-value | | | | | | | | 0.0037 |
| CV (%) | | | | | | | | 42.45 |

Notes:
Figures in brackets were average damage scores for the different treatments.
Treatment means followed by different letters are statistically significantly different at $P > 0.05$

At both sites, amitraz was the most effective acaricide against the tobacco mite being statistically significantly more effective than malation and dimethoate. On the other hand, dimethoate was the least effective although it was not statistically significantly less effective than malathion and abamectin.

### 4.2.2 Effects of acaricides on vegetable natural enemies under field conditions.

Only general observations were made on the effects of acaricides on vegetable natural enemies in the field. At establishment many different species of different natural enemies including ladybird beetles, hoverflies, spiders, predatory wasps and *Oligota* species were seen in both sites. Higher incidences of natural enemies were noticed from establishment when aphids and whiteflies attacked the crop. However, when the broad-spectrum insecticide carbarly was used against the aphids, whiteflies and bollworms, there was a marked decrease in the incidences of these natural enemies. When acaricide sprays were finally started, the incidences of natural enemies decreased further. Adult hoverflies, spiders and wasps were seen flying and crawling (spiders) around. No hoverfly larvae were found in the plots. None of the natural enemies were found dead in the field.

# CHAPTER FIVE

# DISCUSSION

## 5.1 Resistance status of the tobacco spider mite

Laboratory bioassay results indicated that the tobacco spider mite populations on tomatoes from both Mutoko and Chinamhora were beginning to develop resistance against the currently recommended and most used acaricides. The Mutoko population was more resistant to dimethoate than the Chinamhora population. This could be because of the fact that Mutoko farmers had been using dimethoate, an organophosphate acaricide, for a longer period than those in Chinamhora. The longer the period a pest is exposed to a pesticide the faster it develops resistance against it. It could also have been due to the fact that Chinamhora farmers, by their proximity to the city of Harare, had more access to other acaricides thus, reducing the period of dimethoate exposure to the pest. The fact that the tobacco mite populations from both areas were already showing signs of resistance against abamectin, a relatively new chemical, was of great concern.

The acaricide field efficacy results from the two sites in Chinamhora collaborated the laboratory results. That is, of the four acaricides tested, dimethoate was the least effective acaricide against the tobacco mite at the recommended rates. In other words, its ineffectiveness could only be ascribed to resistance since the water used for spray mixture preparation and the method of application were more or less similar for all four acaricides.

Based on the current results, there is an urgent need to put in place strategies to delay or slow down further development of resistance in *T. evansi* in tomatoes. This however could have been happening already as farmers were using a wide range of insecticides and/or acaricides, though not as a strategy for resistance management but because they were trying to avoid high costs of some of these chemicals and also due to non-availability of the same chemicals all the time. Some could not even afford to spray and so grew tomatoes during the cool periods (Dagnoko and Kwaramba 1999).

It was possible that part of the mite population in one location was left untreated keeping the susceptible genes in the pool. There was probably already an acaricide in use in Chinamhora which, like abamectin, acted by stimulating the release of γ-aminobutyric acid inhibiting the neurotransmitters and causing paralysis in motile stages of mites. This situation is known to occur where arthropods developed resistance to new yet unused chemicals (Denholm, 1992).

The four acaricides tested fall into three chemical groups of compounds i.e. organophosphates (dimethoate and malathion), avermectins (abamectin) and amidines (amitraz). Since the mode of action of these compounds against pests such, as spider mites are different, the mechanisms of resistance by the tobacco mite against these acaricides are bound to be also different. There is evidence however that the resistance mechanism towards organophosphates (dimethoate and malathion) could have been behavioural according to Duncombe (1976). In support of Duncombe's assertion, Pedigo (1996) and Dirske and Van de Vrie (1996) have reported that mites have also been

reported to avoid areas sprayed with pyrethroids. The question is whether behavioural resistance represents heritable shifts in behaviour or simply survival for a long enough period to exhibit avoidance behaviour.

## 5.2 Effects of selected acaricides on the selected natural enemies of vegetable pests

Current laboratory bioassay results confirmed what other authorities showed. That is, pesticides were generally harmful to natural enemies (Duncombe, undated; Pedigo, 1996; Verkerk, 2001). However, the ladybird beetle had a higher tolerance to malathion and dimethoate than the other natural enemies, as indicated earlier by Verkerk (2001). The results indicated that all the four acaricides were harmful to the ladybird beetles, aphid wasp parasitoid and the predatory mite. This could have been because all the acaricides had some contact mode of action. Due to the harder outer wings (elytra), the ladybird beetle had limited lethal contact compared to the other soft-bodied natural enemies for example the aphid wasp parasitoid.

No natural enemies were found dead in the field. It was possible that they died outside the field or they avoided coming into the sprayed fields. The lethal effects of the four selected acaricides on the natural enemies of vegetable pests under field conditions that were meant to confirm the laboratory studies were not made because no quantitative assessments were made in the field studies.

## 5.3 Resistant Pest Management

Pesticide resistance in *T. evansi* seems a key looming constraint to tomato production in Zimbabwe. Curative resistance management presents major challenges for farmers, scientists and the agrochemical industry. As there is evidence of resistance development, it is necessary to develop a package of commonsense management practices that can be easily followed by farmers to make efficient use of available acaricides while research is being done on more environmentally friendly methods of control as has happened before in cotton (Duncombe, unpublished and undated). Based on these general observations and current research results, it is clear that insecticide resistance management (IRM) and integrated pest management (IPM) strategies should be implemented/developed as short- and long-term solutions, respectively, to forestall resistance development in the tobacco mite on tomatoes.

### 5.3.1 IRM

The IRM strategies to be implemented should be targeted at those operational factors that have been identified as promoting the development of resistance in the tobacco mite on tomatoes. Taylor and Georghio (1979) have categorized these factors as chemical and application.

#### 5.3.1.1Chemical factors

Among the chemical factors, there is no doubt that the tobacco mite has been exposed to dimethoate, a broad-spectrum organophosphate insecticide, for a long time. Currently, it is also being exposed to malathion, also an organophosphate which is closely related to

dimethoate. In addition, the mite is also exposed to pyrethroids and orgachlorines used to control the African bollworm, *Helicoverpa armigera* which also attacks tomatoes. Based on these factors, the following IRM strategies are suggested:

- Selective acaricides like tetradifon should be used
- Pyrethroids known to promote mite reproduction e.g. cypermethrin, should be avoided
- Rotation of acaricides that do not have the same mode of action should be encouraged
- If mixtures are required, only pesticides, which do not have the same mode of action should be mixed.

Acaricide rotations using different chemical groups will delay or slow down the development of resistance (Grout *et al*, 1998). The greater the number of products used with different modes of action the longer it may take before strains develop resistance to any of the products. Rotating dimethoate or malathion with mitac followed by abamectin in time and/or space would help. It is important to adhere to the rotation programme (Dirkse and Van de Vrie, 1996). This strategy however assumes that the resistant individuals are less fit when there is no selection pressure and that there is no multiple resistance between the pesticides used (Grout *et al*, 1998; Brettell, 1995). Rotations both in space and time have worked before in cotton.

Use of mixtures of compatible acaricides that belong to different groups will also delay development of resistance (Pedigo 1996; Grout *et al* 1998). This strategy is useful against

pests of high value crops and medical pests but involves costs, imposes environmental risks, residues, destruction of natural enemies and a risk of 'super' resistance (resistance to several compounds at once). This will reduce the tendency for mites not controlled by the single chemical to become dominant and difficult to control. Farmers should however avoid mixing pesticides with the same mode of action (e.g. dimethoate and malathion) as resistance to more than one product may quickly develop. In this case, a mixture of mitac and dimethoate or malathion is effective or mixing abamectin with any of the selected acaricides. Results are, however variable as there is chance of the mites developing cross-resistance between groups of the pesticides.

#### 5.3.1.2 Application factors

Currently, there are no thresholds for the chemical control of the tobacco mite on tomatoes, which means there is generally frequent use of pesticides. This in turn means that every generation of this pest is selected since it has a relatively short generation time. The fact that most of the acaricides currently recommended are systemic e.g. dimethoate and amitraz, means that no functional refugia exist as coverage by the systemic acaricides is effectively complete so that no part of the population remains unselected. Since tomatoes are grown all year round, it means that all population generations are likely to be treated. Based on the above application factors, the following IRM strategies are suggested:

- The first acaricide spray should be applied as soon as the pest is recorded and subsequent sprays when there is a substantial rise in the pest level compared with the previous record

- Spot sprays rather than overall sprays should be applied
- Alternation of acaricides in time and/or space at farm level should be practiced.

Saturation i.e. the application of high doses than recommended is applicable to high-value crops and medical pests but like pesticide mixtures has adverse impact on the environment. The insect defense mechanism is saturated by doses that overcome resistance by applying higher dosages on target to render the resistance gene functionally recessive resulting in a decrease in the rate of resistance development. This tactic may not be practical if some selection has already begun. Detoxification mechanisms may be suppressed by use of synergists that function by inhibiting specific detoxification enzymes. However, some insects have been reported to develop resistance to insecticide-plus-synergist mixtures (Pedigo, 1996). This has not been reported in *T. evansi*. Saturation delays the onset of resistance to chemicals by pests. For saturation use of short residual pesticides especially those with systemic action against active stages of mites is recommended (Grout *et al*, 2000: Pedigo, 1996).

Alternation of pesticides is equivalent to geographic rotation where different unrelated pesticides are used in different parts of the same fields or parts of the country from year to year (Grout *et al* 1998; Duncombe 1976). The country can be subdivided into zones and each zone uses a different set of acaricides as has been done in cotton in Zimbabwe. Alternatively, farmers themselves can subdivide their fields and do the same at farm level. Another chemical will kill mites not killed by one chemical when they migrate.

According to Pedigo (1996), this is more effective against less mobile pests like the tobacco spider mite (*T. evansi*).

### 5.3.2 IPM

IPM has been defined by Pedigo (1996) as the management of pests that emphasize rational use of pesticides to conserve natural enemies in agro-ecosystems. Conservation of natural enemies can also be achieved by using non-chemical pest management tactics such as ecological management, host plant resistance and biological control. Only in those cases where non-chemical management tactics cannot cope, only then, can the rational use of pesticides employed as a last resort.

Use of natural enemies has been implicated as a key factor in reducing spider mite populations in many agricultural and horticultural systems. Disruption of beneficial populations including predators by acaricides has been cited as a cause of spider mite outbreaks (Trichilo and Wilson, 1993; Gurr *et al*, 1996). A number of spider mites are known to feed on spider mites. These include predatory mites, ladybird beetles, lacewings, predatory thrips, mired bugs, and cecidomyid and syrphid flies (Verela and Seif, 2000). The predacious mite, *Phytoseilus persimilis* (Athias-Henriot) and *Amlyseus andersoni* (Chant) have been widely used to control *T. urticae* in glasshouse crops in Europe and America (Verela and Seif, 2000). However successes in outdoor crops have been limited to strawberries in California (Verela and Seif, 2000). While several species of indigenous predatory mites are known to occur in eastern and southern Africa, no detailed investigations in their role in tomato fields has yet been conducted (Varela and

Seif, 2000). Up to now only small staphylinid beetles (*Oligota* sp.) are known to feed on *T. evansi*, and efforts are on the way to import predators from South America, the origin of *T. evansi*, and release them in Africa in a classical biological control approach Verela and Seif, 2000)

### 5.3.2.1 Chemical strategies

Rational use of pesticides means that:

- Pesticide application should be based on scouting and pre-determined thresholds
- Selective pesticides (physiological selectivity) should be used
- Short persistent pesticides should be used
- Low pesticide dosages should be applied
- Selective placement of pesticides (ecological selectivity) should be practiced

According to Dent (1993), insecticides have always been the backbone of insect pest control since the early 1950s. The most common error farmers make is to withhold spraying when spider mite population is still low. To be fully effective, chemical control measures should commence as soon as spider mites are detected. Delaying the initially acaricide application results in an increase in mite population making it more difficult to control once spraying is eventually started (Pedigo, 1996). It must be noted that spraying too early leads to unnecessary use of insecticides, whereas spraying too late may lead to significant losses in quality and quantity of produce as well as greater risk of resistance developing.

Preventive applications and applications of dosages lower than recommended must be avoided for mites. Use of the appropriate doses and times of application are necessary. A thorough understanding of the different chemical formulations and their mode of action is necessary. Good overall spray coverage is very important and a prerequisite for chemical control especially where contact acaricides are employed. This is difficult as mites become established under the leaf, and form the typical protective web. Use of functional nozzles and spraying equipment is necessary. It has been reported in several surveys and observations that equipment used by smallholder farmers is usually non-functional.

The indiscriminate use of non-selective broad-spectrum insecticides, especially pyrethroids and organophosphates which are detrimental to natural enemies lead to an increase in the frequency of outbreaks of mites and create conditions favourable for resurgence and/or replacement. Besides, according to Duncombe (1972), it is also necessary to avoid insecticides like dimethoate and related organophosphates like monocrotophos and triazophos that enhance spider mite reproduction. Preservation of refugia by leaving some generations or part of the population untreated, spot spraying hot spots only and making local rather than area wide chemical applications where practical is advisable. As was shown by Matthews (1984) and Verkerk, dimethoate, abamectin, amitraz and malathion were harmful to adult *Phytoseiulus persimilis*. There was need therefore to use chemicals that were not broad-spectrum in action but very specific on the target pest. This would conserve natural enemies and this would help in the development

and implementation of an IPM programme. With the whole world moving towards lowering pesticide use on food crops, this would be a move tomato growers would adopt.

#### 5.3.2.2 Non-chemical strategies

The following non-chemical management tactics should be integrated with the rational use pesticides outlined above.

General hygiene becomes very important in lowering populations of mites. Routine preventive control practices such as preventing mites from going to clean fields; destruction of other hosts etc should be routinely practiced to minimize development of resistance. Separate infected crops from newly planted crops or nursery areas. During tomato production, favourable conditions namely hot, dry weather for red spider mite reproduction should be avoided where possible. Instead, late summer and late winter can be suitable times but care must be taken in areas where night frost forms another threat to the crop. Infested crops and other hosts should be burnt or removed during the season. Reducing plant distances and applying overhead irrigation influences the microclimate and represses the mite populations. Care must be taken as this enhances fungal diseases. Water and nutrient stress must be avoided as this increases mite populations. Mulching and addition of organic matter to the soil improves water-holding capacity and decreases evaporation. Workers must not move from infested fields to less infested or uninfected plantings to prevent pests from being carried on clothing from one field into another. Finally, crop rotation in order to prevent a build up of the hard-to-kill mites is necessary.

# CHAPTER SIX

# CONCLUSION AND RECOMMENDATIONS

## 6.1 Conclusion

All the acaricides that were tested had RF values that were $\geq 1.0$. Although the RF values were low, indications were that the toacco mite was developing resistance especially to dimethoate. Tests with the same acaricides indicated that they were all harmful to natural enemies commonly found in vegetables. Some aspects of both IRM and IPM were identified as short- and long-term strategies respectively, for forestalling or delaying resistance development in the tobacco spider mite in tomatoes.

## 6.2 Recommendations

Based on current investigations, the following IRM and IPM strategies were recommended in order to delay and/or avert resistance development in the tobacco mite in tomatoes.

### 6.2.1 IRM

#### 6.2.1.1 Strategies to anilate chemical factors

- Selective acaricides like tetradifon should be used
- Pyrethroids known to promote mite reproduction e.g. cypermethrin, should be avoided
- Rotation of acaricides that do not have the same mode of action should be encouraged

- If mixtures are required, only pesticides, which do not have the same mode of action should be mixed.

#### 6.2.1.2 Strategies to anilate application factors

- The first acaricide spray should be applied as soon as the pest is recorded and subsequent sprays when there is a substantial rise in the pest level compared with the previous record
- Spot sprays rather than overall sprays should be applied
- Alternation of acaricides in time or space at farm level should be practiced.

### 6.2.2 IPM

#### 6.2.2.1 Chemical strategies

- Pesticide application should be based on scouting and pre-determined thresholds
- Selective pesticides (physiological selectivity) should be used
- Short persistent pesticides should be used
- Low pesticide dosages should be applied
- Selective placement of pesticides (ecological selectivity) should be practiced

#### 6.2.2.2 Non-chemical strategies

- Alternative host plants should be destroyed
- If possible, tomatoes should be grown in late summer or winter after the danger of frost has passed

- Nutrient and water stress of the crop should be avoided
- Workers must not move from infested fields to less infested or un-infested fields or work from old fields to new fields
- Crop rotation should be practiced to prevent a build-up of the pest

## REFERENCES


**Anonymous, 2000.** Conserving natural enemies. Working with natural enemies series. Bulletin No 2. CABI Bioscience. Berkshire. UK. pp34-46

**Anonymous, 2001.** Zimbabwe Crop Chemical Handbook. Vol.1 Insecticides, Fungicides, Growth regulants, Fumigants and Health products. ACIA/Department of Research and Specialist Services. Harare. Zimbabwe. pp 13

**Borror, D. J.; Triplehorn, C. A. and Johnson, N. F. 1982.** An introduction to the study of insects. 6th edition. Saunders College Publishing. Harcourt Brace Jovanovich College publishers. New York. USA. Pp 10

**Brettell, J. H. and Burgess, M. W. 1971.** A preliminary assessment of the effects of some insecticides on predators of cotton pests. Rhodesian Agric. J., 70(5) Bulletin 2641. pp 103-104. pp-8

**Brettell, J. H. 1979.** Green lacewings (Neuroptera: Chrsopidae) of cotton fields in central Rhodesia. 1. Biology of *Chrysopa boninensis* Okomoto and toxicity of certain insecticides to the larva. Rhodesia Journal of Agricultural Research. 17: 141-150. pp 3

**Brettell, J. H. 1984**. Green lacewings (Neuroptera: Chrysopidae) of cotton fields in Zimbabwe. 3 Toxicity of certain acaricides, aphicides and pyrethroids to larvae of *Chrysopa boninensis* Okamoto *Chrysopa congrua* Walker Okamoto and *Chrysopa pudica* Navas. Zimbabwe Journal of Agricultural Research. 22: pp 133-139. pp 7

**Brettell, J. H. 1995.** Pest management strategies and decision making in Zimbabwe. Cotton Training Centre. Kadoma Zimbabwe. pp57

**Cremyln, R. 1978**. Pesticides: Preparation and mode of action. John Wiley & Sons. Chichester. Pp 1

**Dagnoko, M. and Kwaramba, J. R. 1999**. Review of vegetable IPPM potential in Zimbabwe: A case study of Mashonaland East Province. Unpublished. Pp 12-54

**Dent, D. 1991**. Insect pest management. CABI. Wiltshire. UK. Pp 18-56

**Dirkse, F.B. and Van de vrie, M. 1996**. Managing Pesticide Resistance in Greenhouses. Horticultural Quarterly View. October issue. pp 14-57.

**Dobson, H., Cooper, J., Manyangarirwa, W., Karuma, J. and Chiimba, W. 2002.** Integrated Vegetable Pest Management. NRI/ DR&SS. Pp 11

**Dube, Bek.; Dube, Bong.; Gova, M., Makaya, P. R., Mutimutema, E., Savoure~, P., Letheve~, X., and Turner, A. 1996.** Important vegetable pests and diseases in Zimbabwe. Identification and Control. Agritex, CIIFAD, Cooperation France Zimbabwe, Strand Multiprint. USA. Pp 15

**Duncombe, W. G. 1970.** Acaricide screening techniques in use in Rhodesia. Rhodesia Science News. Vol. 4, No. 3. pp 99-100 pp 27

**Duncombe, W. G. 1972.** *Tetranychid* Mites as pests in Cotton. Department of Research and Specialist Services. Salisbury. Rhodesia. Pp 7-57

**Duncombe, W. G. 1976.** Cotton losses caused by spider mites- (Acarina: Tetranychidae). Rhodesia Agric. Journal. Vol 74 (6). Pp 3-58

**Duncombe, W. G. Undated.** The acaricide spray rotation for cotton. Cotton Research Institute. Gatooma. Rhodesia pp 20

**Finney, D. J., 1962.** Probit analysis- A statistical treatment of the sigmoid response curve. Second edition. Cambridge University Press. Great Britain.pp 35

**Grout T.G,; Hofmeyr, J. H. Hattingh, V. Buitendag, C.H. and Ware, A. B. 1998.** Integrated pest management. In: Integrated pest and disease management. Volume III. Research and Extension Services. Outspan International Ltd. Centurion. South Africa. Pp 7-59

**Hussey, N. W., and Scopes, N. E. A. 1985.** Damage and control. Greenhouse vegetables. Britain. In: Spider mites. Their biology, natural enemies and control. Vol. 1B Ed. Melle, M. and Sabelis, M. W., Eds. Elsevier, Amsterdam, 285-97. pp 32-38

**Kochhar, S. L.1986.** Tropical Crops- a textbook of economic botany. MacMillan Publishers Ltd. Hong Kong. Pp 1-6

**Magdalena, K. P. and Meyer S. 1996.** Mite pests and their predators on cultivated plants in Southern Africa: vegetables and berries. Plant Protection Research Institute. Pretoria. South Africa. Pp 21

**Magdalena, K. P. and Meyer S. 1981.** Mite pests of crops in Southern Africa. Science Bulletin N0. 397. Plant Protection Research Institute. Pretoria. South Africa. Pp 8-11

**Mark, J. and Kidd, N. 1996.** Insect natural enemies. Chapman and Hall. London. UK. Pp 2

**Matthews 1984.** Pest management. Longman Group Ltd. New York pp 14-52

**Munthali, D. C. 1991**. Biological efficiency of small pesticide deposits. In. Dent, D. Insect pest management. CABI. Wiltshire. UK. Pp 18-21

**Musundire, R. 2002**. Evaluation of certain aspects of chemical varietal and biological control methods against broad bean (*Vicia faba*) leaf miners (Liliomyza huidobriensis). University of Zimbabwe Harare. Zimbabwe. pp 28.

**Pedigo, L. P. 1996**. Entomology and pest management. Second edition. Prentice Hall Inc. New Jersey. USA. Pp 1-59

**Sawicki, R. M. and Denholm, I. 1987**. Management of resistance to pesticides in cotton pests. Tropical Pest management 33(4) 262-272. pp 2-51

**Tag El-Din, M. H. 1992**. Development of resistance to organophosphates and carbamates in *Tetranychus urticae* Koch in fruit orchards. Acta. Phytopathologica et Entomologica Budapest. Hungarica. Vol 62 (5) Pp. 605-608. pp 16-18

**Taylor, E. D. 1981**. Red spider mite. Zimbabwe Agric. J. Vol. 78 (2) pp 69-70. pp 16

**Taylor, C. E. and Georghio, G. P. 1979**. Suppression of insecticide resistance by alteration of gene dominance and migration. Journal of economic entomology. Vol. 72 pp105-9. pp 15-17

**Turner, A. Chivinge, O.1999**. Production and marketing of horticultural Crops in Zimbabwe: A survey of smallholder farmers in the Mashonaland East Province. Cornell International Institute for Food Agriculture and development. (CIIFAD) Ithaca. New York. USA. Pp 1-4

**Varela, A. M. and Sief, A. A. 2000**. Pest Management Manual for tomatoes. GTZ-IPM Horticulture project. ICIPE. Nairobi. Kenya. Pp 5-23

**Verkerk, R. 2001**. Farmers' Friends: Recognition and conservation of natural enemies of vegetable pests. NRI/ DR&SS. Pp 2-52

**Wells, P. D. 1980**. Tomato Production. Department of Conservation and Extension. Government Printers. Harare. Zimbabwe. pp 4

**Worthing, C. R. and Walker, S. (Editors). 1983**. The Pesticide Manual – A World Compendium. Seventh edition. The British Crop Protection Council. Pp30

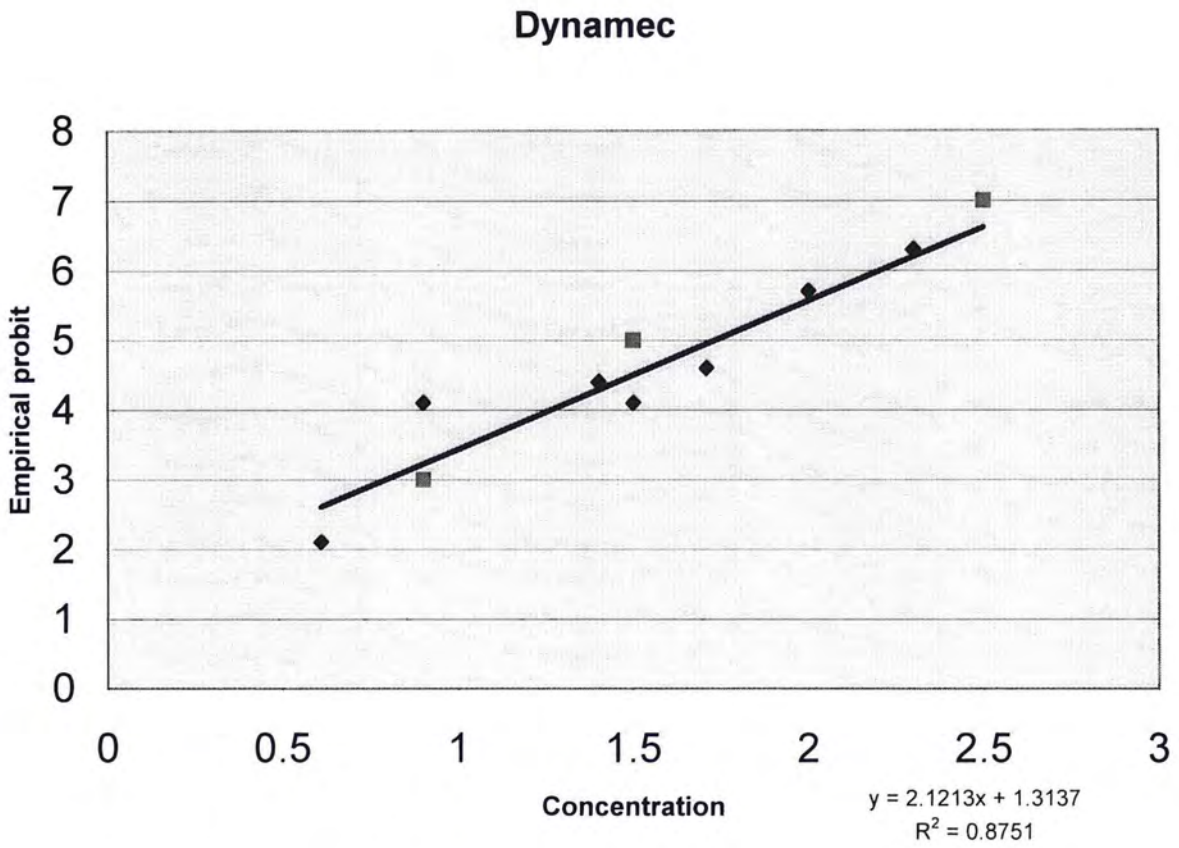


Fig 4.1 Regression line for dynamec

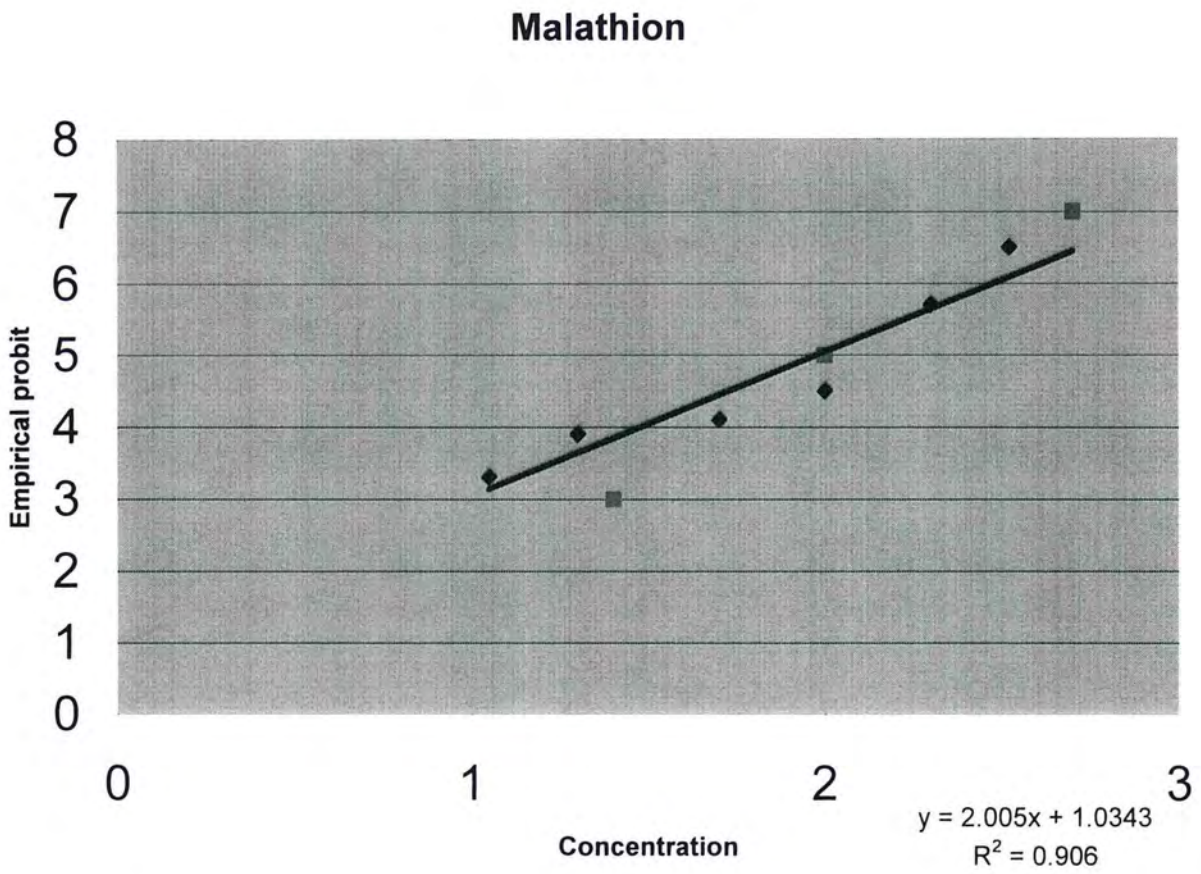


Fig. 4.2 Regresion line for malathion

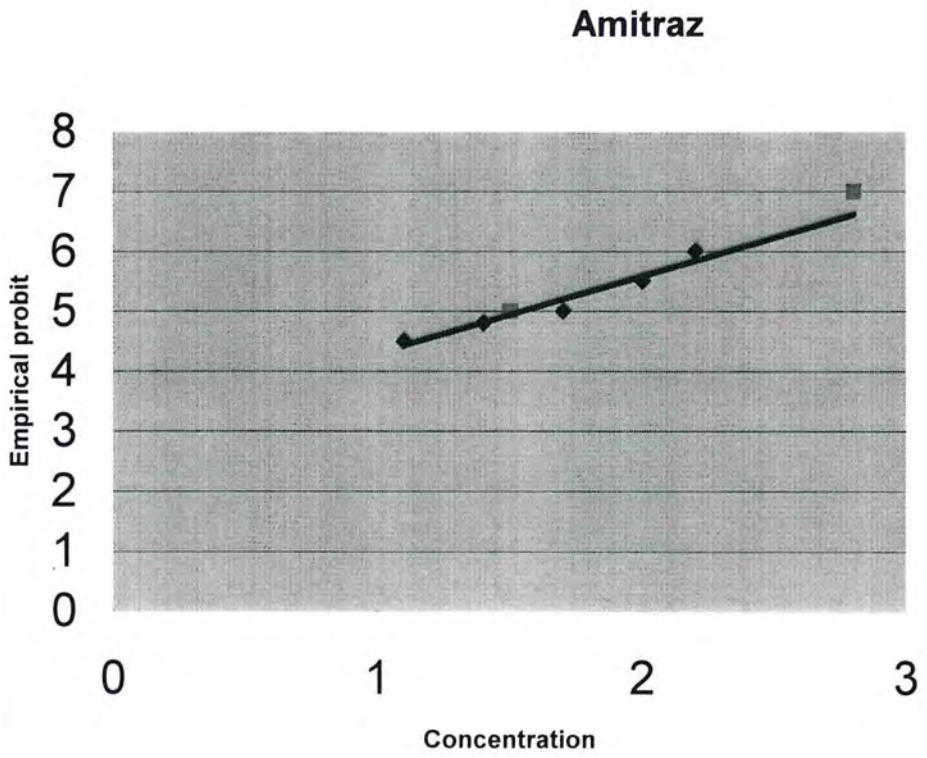


Fig 4.3 Regression line for amitraz

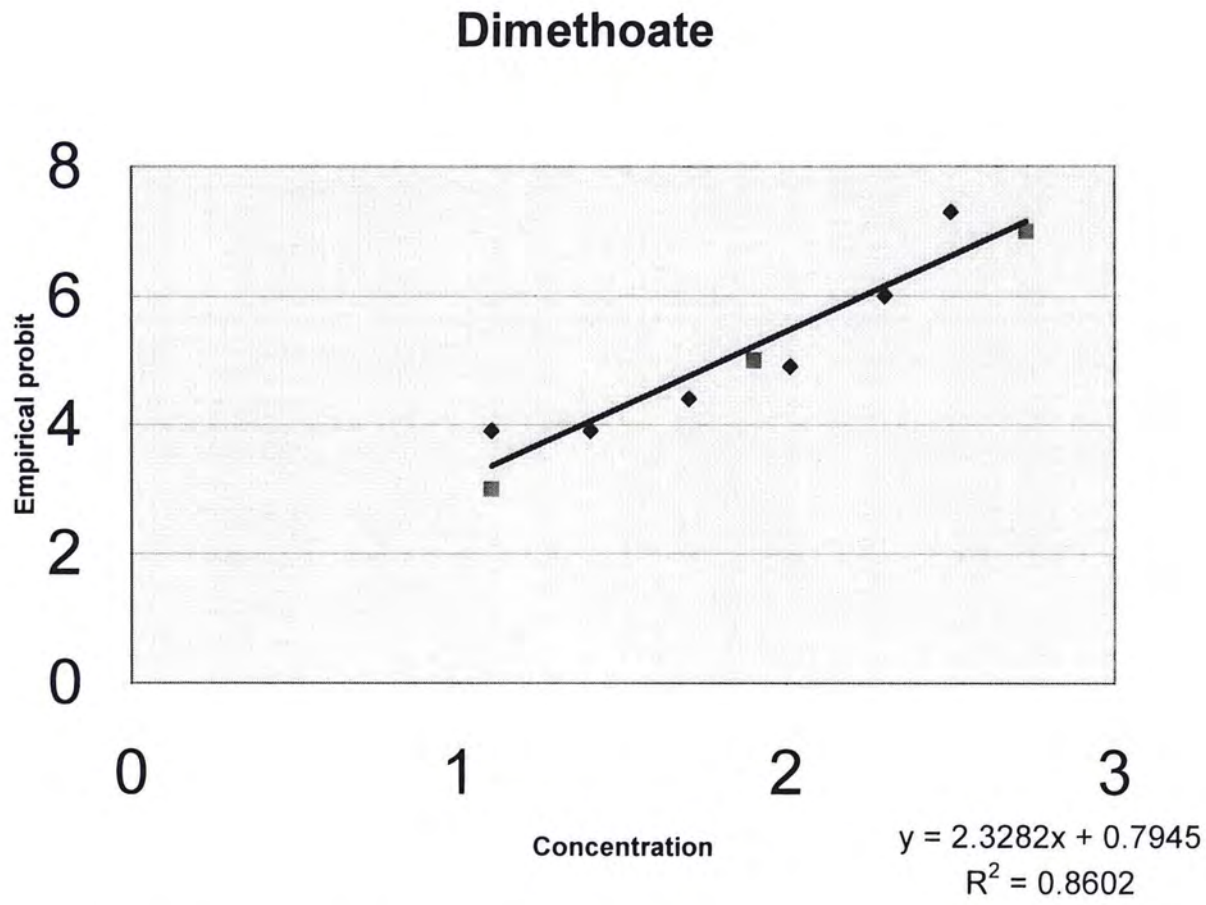


Fig. 4.4 Regresion line for dimethoate

**Appendix 1 Lethal concentrations for the susceptible strain of tobacco spider mites**

**1a Abamectin**

| gai/l | Hours | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Total mites dead | Actual % mortality | Corrected % mortality |
|---|---|---|---|---|---|---|---|---|---|
| 0.0234 | 24 | 2 | 6 | 5 | 7 | 6 | | | |
| | 48 | 10 | 10 | 10 | 10 | 10 | 50 | 100 | 100% |
| | 72 | | | | | | | | |
| | 96 | | | | | | | | |
| 0.0117 | 24 | 3 | 0 | 0 | 0 | 0 | | | |
| | 48 | 3 | 1 | 1 | 1 | 1 | | | |
| | 72 | 8 | 2 | 4 | 3 | 1 | | | |
| | 96 | 9 | 7 | 7 | 8 | 8 | 39 | 78 | 76.6 |
| 0.00585 | 24 | 0 | 2 | 0 | 0 | 0 | | | |
| | 48 | 0 | 2 | 1 | 0 | 0 | | | |
| | 72 | 3 | 2 | 3 | 1 | 1 | | | |
| | 96 | 6 | 3 | 6 | 3 | 2 | 20 | 40 | 36.2 |
| 0.002925 | 24 | 0 | 0 | 0 | 0 | 0 | | | |
| | 48 | 1 | 0 | 0 | 1 | 0 | | | |
| | 72 | 2 | 3 | 0 | 2 | 0 | | | |
| | 96 | 2 | 4 | 3 | 2 | 4 | 15 | 30 | 25.5 |
| 0.0014625 | 24 | 0 | 0 | 0 | 0 | 1 | | | |
| | 48 | 1 | 0 | 0 | 0 | 1 | | | |
| | 72 | 1 | 0 | 1 | 0 | 2 | | | |
| | 96 | 2 | 2 | 3 | 2 | 3 | 12 | 24 | 19.1 |

**Appendix 1b Malathion**

| gai/l | Hours | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Total mites dead | Actual % mortality | Corrected % mortality |
|---|---|---|---|---|---|---|---|---|---|
| 1.89 | 24 | 6 | 7 | 8 | 4 | 5 | | | |
| | 48 | 10 | 9 | 10 | 7 | 7 | | | |
| | 72 | 10 | 10 | 10 | 9 | 8 | | | |
| | 96 | 10 | | 10 | 10 | 10 | 50 | 100 | 100 |
| 1.26 | 24 | 4 | 3 | 1 | 4 | 3 | | | |
| | 48 | 6 | 5 | 4 | 7 | 5 | | | |
| | 72 | 8 | 7 | 7 | 8 | 5 | | | |
| | 96 | 8 | 9 | 7 | 8 | 6 | 38 | 76 | 74.5 |
| 0.63 | 24 | 3 | 1 | 0 | 1 | 0 | | | |
| | 48 | 3 | 1 | 1 | 1 | 0 | | | |
| | 72 | 4 | 2 | 1 | 3 | 1 | | | |
| | 96 | 6 | 3 | 1 | 4 | 3 | 17 | 34 | 29.8 |
| 0.315 | 24 | 0 | 0 | 0 | 1 | 1 | | | |
| | 48 | 0 | 0 | 2 | 1 | 1 | | | |
| | 72 | 2 | 3 | 2 | 1 | 1 | | | |
| | 96 | 2 | 5 | 2 | 1 | 2 | 12 | 24 | 19.1 |
| 0.158 | 24 | 1 | 1 | 0 | 0 | 0 | | | |
| | 48 | 1 | 1 | 1 | 0 | 0 | | | |
| | 72 | 1 | 1 | 2 | 1 | 1 | | | |
| | 96 | 2 | 2 | 2 | 1 | 2 | 9 | 18 | 12.8 |
| 0.079 | 24 | 0 | 0 | 2 | 0 | 0 | | | |
| | 48 | 1 | 0 | 2 | 0 | 0 | | | |
| | 72 | 2 | 0 | 2 | 0 | 0 | | | |
| | 96 | 2 | 1 | 2 | 0 | 0 | 5 | 10 | 4.3 |

**Appendix 1c: Amitraz**

| gai/l | Hours | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Total mites dead | Actual % mortality | Corrected % mortality |
|---|---|---|---|---|---|---|---|---|---|
| 1.2 | 24 | 3 | 2 | 2 | 3 | 5 | | | |
| | 48 | 5 | 6 | 4 | 7 | 7 | | | |
| | 72 | 7 | 10 | 8 | 10 | 9 | | | |
| | 96 | 9 | 10 | 10 | 10 | 10 | 50 | 100 | 100 |
| 0.6 | 24 | 1 | 0 | 0 | 0 | 1 | | | |
| | 48 | 2 | 0 | 2 | 1 | 1 | | | |
| | 72 | 5 | 3 | 4 | 5 | 6 | | | |
| | 96 | 8 | 7 | 6 | 7 | 8 | 36 | 72 | 70.2 |
| 0.3 | 24 | 0 | 0 | 0 | 0 | 1 | | | |
| | 48 | 1 | 2 | 2 | 0 | 1 | | | |
| | 72 | 4 | 4 | 3 | 2 | 3 | | | |
| | 96 | 7 | 7 | 5 | 4 | 3 | 26 | 52 | 48.9 |
| 0.15 | 24 | 0 | 0 | 2 | 0 | 1 | | | |
| | 48 | 1 | 0 | 2 | 0 | 0 | | | |
| | 72 | 3 | 0 | 3 | 1 | 2 | | | |
| | 96 | 4 | 3 | 6 | 5 | 5 | 23 | 46 | 42.6 |
| 0.075 | 24 | 0 | 0 | 1 | 0 | 1 | | | |
| | 48 | 0 | 0 | 1 | 0 | 1 | | | |
| | 72 | 1 | 3 | 2 | 1 | 1 | | | |
| | 96 | 6 | 4 | 3 | 3 | 2 | 18 | 36 | 31.9 |

**Appendix 1d Dimethoate**

| gai/l | Hours | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Total mites dead | Actual % mortality | Corrected % mortality |
|---|---|---|---|---|---|---|---|---|---|
| 0.9 | 24 | 8 | 7 | 5 | 6 | 7 | | | |
| | 48 | 10 | 9 | 8 | 8 | 8 | | | |
| | 72 | | 10 | 10 | 9 | 8 | | | |
| | 96 | | | | 10 | 10 | 50 | 100 | 100 |
| 0.6 | 24 | 2 | 6 | 4 | 3 | 5 | | | |
| | 48 | 5 | 6 | 7 | 6 | 7 | | | |
| | 72 | 7 | 7 | 8 | 8 | 7 | | | |
| | 96 | 7 | 9 | 8 | 9 | 9 | 42 | 84 | 83 |
| 0.3 | 24 | 0 | 1 | 1 | 0 | 0 | | | |
| | 48 | 0 | 1 | 2 | 1 | 1 | | | |
| | 72 | 4 | 5 | 3 | 5 | 3 | | | |
| | 96 | 4 | 7 | 3 | 7 | 3 | 24 | 48 | 44.7 |
| 0.15 | 24 | 0 | 0 | 1 | 0 | 0 | | | |
| | 48 | 1 | 1 | 1 | 0 | 0 | | | |
| | 72 | 1 | 3 | 2 | 4 | 4 | | | |
| | 96 | 2 | 3 | 2 | 4 | 4 | 15 | 30 | 25.5 |
| 0.075 | 24 | 0 | 0 | 0 | 0 | 1 | | | |
| | 48 | 0 | 0 | 0 | 0 | 1 | | | |
| | 72 | 0 | 0 | 1 | 1 | 3 | | | |
| | 96 | 1 | 2 | 2 | 1 | 3 | 9 | 18 | 12.8 |
| 0.038 | 24 | 1 | 0 | 0 | 0 | 0 | | | |
| | 48 | 1 | 0 | 1 | 0 | 0 | | | |
| | 72 | 2 | 1 | 1 | 1 | 2 | | | |
| | 96 | 3 | 1 | 1 | 2 | 2 | 9 | 18 | 12.8 |

**Appendix 1e Control (Distilled water)**

| gai/l | Hours | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Total mites dead | % mortality |
|---|---|---|---|---|---|---|---|---|
| 0% | 24 | 0 | 1 | 0 | 0 | 0 | | |
| | 48 | 0 | 1 | 0 | 0 | 0 | | |
| | 72 | 0 | 1 | 0 | 0 | 0 | | |
| | 96 | 0 | 1 | 1 | 0 | 1 | 3 | 6 |

## Appendix 2 Calculation of log-dose/probit regression line

### 2a:Abamectin

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose gai/l | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+3.9) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 0.0234 | 50 | 50 | 100 | 100 | 2.3 | 7.33 | 6.1 | 7.1 | 0.218 | 10.9 | 25.07 | 77.39 | 6.6 |
| 0.0117 | 50 | 39 | 78 | 76.6 | 2 | 5.74 | 5.7 | 5.72 | 0.477 | 23.85 | 47.7 | 136.42 | 5.8 |
| 0.00585 | 50 | 20 | 40 | 36.2 | 1.7 | 4.64 | 4.8 | 4.65 | 0.52 | 26 | 44.2 | 120.9 | 5.0 |
| 0.00293 | 50 | 15 | 30 | 25.5 | 1.4 | 4.36 | 3.9 | 4.45 | 0.247 | 12.35 | 17.29 | 54.96 | 4.0 |
| 0.00146 | 50 | 12 | 24 | 19.1 | 1.1 | 4.12 | 3.1 | 5.57 | 0.038 | 1.9 | 2.09 | 10.58 | 3.5 |
| 0.0% | 50 | 3 | 6 | | | | | | | | | | |

Corrected mortality = $\frac{\text{\% killed in treatment - \% killed in control}}{\text{100 - \% killed in control}}$ X 100

$\sum w = 75$; $\sum wy = 400.25$; $\sum wx = 136.35$; $\sum wxy = 744.94$; $\sum wx^2 = 54.71$; $\sum wy^2 = 2195.48$;

$\bar{x} = \sum wx / \sum w = 1.82$; $\bar{y} = \sum wy / \sum w = 5.34$;

$b = \frac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 2.52$; Regression line: $y = \bar{y} + b(x - \bar{x})$ When y=5 , x=1.7; when y=7 x=2.5; when y=3 x=.9

Variance = 0.0024; Fiducial limits- $m_1 = 1.6$; $m_2 = 1.8$; $X^2 = 16.6$

**2b Malathion**

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+2.2) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 1.89 | 50 | 50 | 100 | 100 | 2.5 | 7.33 | 6.3 | 6.87 | .306 | 15.3 | 38.25 | 105.11 | 6.36 |
| 1.26 | 50 | 38 | 76 | 74.5 | 2.3 | 5.67 | 5.7 | 5.65 | .477 | 25.85 | 59.46 | 146.05 | 5.76 |
| 0.63 | 50 | 17 | 34 | 29.8 | 2 | 4.48 | 4.9 | 4.49 | .537 | 26.7 | 53.4 | 119.88 | 4.87 |
| 0.315 | 50 | 12 | 24 | 19.1 | 1.7 | 4.12 | 4.1 | 4.13 | .32 | 16 | 27.2 | 66.08 | 3.98 |
| 0.158 | 50 | 9 | 18 | 12.8 | 1.4 | 3.87 | 3.3 | 4.19 | .07 | 3.5 | 4.9 | 14.67 | 3.1 |
| 0.079 | 50 | 5 | 10 | 4.3 | 1.1 | 3.25 | 2.5 | 4.6 | .003 | .15 | .165 | 0.96 | 2.2 |
| 0.0 | 50 | 3 | 6 | | | | | | | | | | |

$\sum w = 87.5$; $\sum wy = 452.48$; $\sum wx = 183.38$; $\sum wxy = 972.13$; $\sum wx^2 = 392.47$; $\sum wy^2 = 2423.10$

$\bar{x} = \sum wx / \sum w = 2.10$; $\bar{y} = \sum wy / \sum w = 5.17$;

$b = \frac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 2.97$; Regression line: $y = \bar{y} + b(x - \bar{x})$; when y=5, x= 2.0 when y=7, x=2.7; when y=3, x= 1.4

Variance = 0.0014; Fiducial limits- $m_1 = 1.93$; $m_2 = 2.07$; $X^2 = 18.67$

## 2c Amitraz

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+2.4) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 1.2 | 50 | 50 | 100 | 100 | 2.3 | 7.05 | 6.0 | 6.65 | .398 | 19.9 | 45.77 | 132.34 | 6.00 |
| 0.6 | 50 | 36 | 72 | 70.2 | 2.0 | 5.52 | 5.6 | 5.53 | .498 | 24.9 | 49.8 | 137.70 | 5.76 |
| 0.3 | 50 | 26 | 52 | 48.9 | 1.7 | 4.97 | 5.2 | 4.97 | .546 | 27.3 | 46.41 | 135.68 | 5.3 |
| 0.15 | 50 | 23 | 46 | 42.6 | 1.4 | 4.82 | 4.8 | 4.81 | .520 | 26 | 36.4 | 125.06 | 4.79 |
| 0.075 | 50 | 18 | 36 | 31.9 | 1.1 | 4.52 | 4.4 | 4.54 | .424 | 21.2 | 23.32 | 96.25 | 4.3 |
| 0.0 | 50 | 3 | 6 | | | | | | | | | | |

$\sum w = 119.3$; $\sum wy = 627.03$; $\sum wx = 201.7$; $\sum wxy = 1091.37$; $\sum wx^2 = 360.38$; $\sum wy^2 = 3354.39$;

$\bar{x} = \sum wx / \sum w = 1.69$; $\bar{y} = \sum wy / \sum w = 5.26$;

$b = \frac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 1.62$; Regression line: $y = \bar{y} + b(x - \bar{x})$, When y=5, x= 1.5; when y=7, x=2.8 when y=3, x=0.23

Variance = 0.0039; Fiducial limits- $m_1 = 1.4$; $m_2 = 1.6$; $X^2 = 4.87$

## 2d **Dimethoate**

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l. | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+2.5) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 0.9 | 50 | 50 | 100 | 100 | 2.5 | 7.33 | 6.45 | 7.01 | .246 | 12.3 | 30.75 | 86.22 | 6.44 |
| 0.6 | 50 | 42 | 84 | 83 | 2.3 | 5.95 | 6.0 | 5.95 | .398 | 19.9 | 45.77 | 118.41 | 5.94 |
| 0.3 | 50 | 24 | 48 | 44.7 | 2 | 4.77 | 5.3 | 4.85 | .540 | 27.0 | 54.0 | 130.95 | 5.20 |
| 0.15 | 50 | 15 | 30 | 25.5 | 1.7 | 4.36 | 4.6 | 4.35 | .480 | 24.0 | 40.8 | 104.4 | 4.46 |
| 0.07 5 | 50 | 9 | 18 | 12.8 | 1.4 | 3.87 | 3.9 | 3.87 | .247 | 12.35 | 17.29 | 47.79 | 3.72 |
| 0.03 8 | 50 | 9 | 18 | 12.8 | 1.1 | 3.87 | 3.2 | 4.36 | .053 | 2.65 | 2.92 | 11.55 | 3.0 |
| 0.0 | 50 | 3 | 6 | | | | | | | | | | |

$\sum w = 98.2$; $\sum wy = 499.32$; $\sum wx = 191.53$; $\sum wxy = 1006.91$; $\sum wx^2 = 386.93$;

$\sum wy^2 = 2633.5$; $\bar{x} = \sum wx / \sum w = 1.95$; $\bar{y} = \sum wy / \sum w = 5.08$;

$b = \dfrac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 2.47$; Regression line: $y = \bar{y} + b(x - \bar{x})$, when y=5, x=1.92; when y=7,x=2.73; when y=3, x=1.10

Variance = 0.0167; Fiducial limits- $m_1 = 1.7$; $m_2 = 2.2$; $X^2 = 14.85$

**Appendix 3 Bioassays for suspected resistant strains**

**3a Chinamhora**

| Treatment | gai/l | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Total | % response (death) | Corrected % mortality |
|---|---|---|---|---|---|---|---|---|---|
| Abamectin | 0.0234 | 9 | 8 | 6 | 7 | 8 | 38 | 76 | 75 |
| | 0.0117 | 5 | 7 | 8 | 6 | 5 | 31 | 62 | 60.4 |
| | 0.00585 | 2 | 1 | 2 | 3 | 1 | 9 | 18 | 14.6 |
| Malathion | 1.89 | 5 | 8 | 7 | 8 | 6 | 34 | 68 | 66.7 |
| | 1.26 | 5 | 7 | 4 | 6 | 5 | 27 | 54 | 52.1 |
| | 0.63 | 2 | 2 | 4 | 3 | 3 | 14 | 28 | 25 |
| Amitraz | 1.2 | 7 | 6 | 8 | 8 | 7 | 36 | 72 | 70.8 |
| | 0.6 | 3 | 3 | 4 | 4 | 3 | 17 | 34 | 31.3 |
| | 0.3 | 2 | 1 | 1 | 2 | 3 | 9 | 18 | 14.6 |
| Dimethoate | 0.9 | 4 | 5 | 4 | 3 | 4 | 20 | 40 | 37.5 |
| | 0.6 | 3 | 2 | 3 | 1 | 2 | 11 | 22 | 18.8 |
| | 0.3 | 0 | 1 | 1 | 1 | 1 | 4 | 8 | 4.2 |
| Control | 0 | 1 | 0 | 0 | 0 | 1 | 2 | 4 | |

**3b Mutoko**

| Treatment | gai/l | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Total | % response (death) | Corrected % mortality |
|---|---|---|---|---|---|---|---|---|---|
| Abamectiin | 0.0234 | 7 | 9 | 9 | 8 | 7 | 40 | 80 | 78.7 |
| | 0.0117 | 6 | 4 | 6 | 5 | 7 | 28 | 56 | 53.2 |
| | 0.00585 | 3 | 3 | 3 | 2 | 2 | 13 | 26 | 21.3 |
| Malathion | 1.89 | 6 | 7 | 8 | 8 | 6 | 35 | 70 | 68.1 |
| | 1.26 | 4 | 6 | 5 | 3 | 3 | 19 | 38 | 34.0 |
| | 0.63 | 2 | 2 | 2 | 3 | 3 | 12 | 24 | 19.1 |
| Amitraz | 1.2 | 7 | 8 | 6 | 6 | 8 | 35 | 70 | 68.1 |
| | 0.6 | 4 | 3 | 4 | 3 | 3 | 17 | 34 | 29.8 |
| | 0.3 | 3 | 2 | 2 | 1 | 2 | 10 | 20 | 14.9 |
| Dimethoate | 0.9 | 7 | 7 | 8 | 8 | 8 | 38 | 76 | 74.5 |
| | 0.6 | 5 | 6 | 6 | 5 | 4 | 26 | 52 | 48.9 |
| | 0.3 | 2 | 1 | 1 | 2 | 1 | 7 | 14 | 8.5 |
| Control | 0 | 1 | 1 | 0 | 0 | 1 | 3 | 6 | |

## Appendix 4 Calculation of log-dose/probit regression lines

### 4a Chinamhora Abamectin

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l. | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+3.9) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 0.0234 | 50 | 38 | 76 | 75 | 2.3 | 5.67 | 5.7 | 5.67 | .532 | 26.6 | 61.18 | 150.82 | 5.9 |
| 0.0117 | 50 | 31 | 62 | 60.4 | 2.0 | 5.25 | 4.9 | 5.3 | .634 | 31.7 | 63.4 | 168.01 | 5.0 |
| 0.00585 | 50 | 9 | 18 | 14.6 | 1.7 | 3.96 | 4.0 | 3.9 | .439 | 21.95 | 37.32 | 85.61 | 4.2 |
| 0.0 | 50 | 2 | 4 | | | | | | | | | | |

$\sum w = 80.25$; $\sum wy = 404.44$; $\sum wx = 161.9$; $\sum wxy = 828.46$; $\sum wx^2 = 330.96$

$\sum wy^2 = 2079.48$; $\bar{x} = \sum wx / \sum w = 2.02$; $\bar{y} = \sum wy / \sum w = 5.04$;

$b = \dfrac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 2.92$; Regression line: $y = \bar{y} + b(x - \bar{x})$, when y=5, x=2.0; when y=7,x=2.7;

Variance= 0.0016; Fiducial limits: $m_1 = 1.92$; $m_2 = 2.08$

## 4b Chinamhora- Malathion

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l. | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+2.2) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 1.89 | 50 | 34 | 68 | 66.7 | 2.5 | 5.44 | 5.4 | 5.43 | .601 | 30.05 | 75.13 | 163.17 | 5.4 |
| 1.26 | 50 | 27 | 54 | 52.1 | 2.3 | 5.04 | 5.0 | 5.06 | .637 | 31.85 | 73.26 | 161.16 | 5.0 |
| 0.63 | 50 | 14 | 28 | 25 | 2 | 4.33 | 4.3 | 4.33 | .532 | 26.6 | 53.2 | 115.18 | 4.4 |
| 0.0 | 50 | 2 | 4 | | | | | | | | | | |

$\sum w = 88.5$; $\sum wy = 439.51$; $\sum wx = 201.59$; $\sum wxy = 1009.02$; $\sum wx^2 = 462.73$;

$\sum wy^2 = 2200.08$; $\bar{x} = \sum wx / \sum w = 2.3$; $\bar{y} = \sum wy / \sum w = 5.0$;

$b = \frac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 2.47$; Regression line: $y = \bar{y} + b(x - \bar{x})$, when y=5, x=1.92; when y=7,x=2.73; when y=3, x=1.10

Variance = 0.00283; Fiducial limits- $m_1 = 2.2$; $m_2 = 2.4$; $X^2 = 14.24$

**4c Chinamhora-Amitraz**

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+2.4) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 1.2 | 50 | 36 | 72 | 70.8 | 2.3 | 5.55 | 5.6 | 5.5 | .558 | 27.9 | 64.17 | 153.45 | 5.43 |
| 0.6 | 50 | 17 | 34 | 31.3 | 2.0 | 4.5 | 4.8 | 4.5 | .627 | 31.35 | 62.7 | 141.08 | 4.68 |
| 0.3 | 50 | 9 | 18 | 14.6 | 1.7 | 3.96 | 4.0 | 3.9 | .439 | 21.95 | 37.32 | 85.61 | 3.93 |
| 0.0 | 50 | 2 | 4 | | | | | | | | | | |

$\sum w = 81.2$; $\sum wy = 380.14$ $\sum wx = 164.19$; $\sum wxy = 780.64$; $\sum wx^2 = 336.43$;

$\sum wy^2 = 1814.74$; $\bar{x} = \sum wx / \sum w = 2.0$; $\bar{y} = \sum wy / \sum w = 4.68$;

$b = \frac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 2.5$; Regression line: $y = \bar{y} + b(x - \bar{x})$, when y=5, x=2.1; when y=7,x=2.9; when y=3, x=1.10

Variance = 0.0023; Fiducial limits- $m_1 = 2.01$; $m_2 = 2.2$; $X^2 = 15.22$

**4d Chinamhora - Dimethoate**

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+2.3) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 0.9 | 50 | 20 | 40 | 37.5 | 2.5 | 4.69 | 4.7 | 4.68 | .616 | 30.8 | 77.0 | 144.14 | 4.66 |
| 0.6 | 50 | 11 | 22 | 18.8 | 2.3 | 4.12 | 4.1 | 4.12 | .471 | 23.55 | 54.17 | 97.03 | 4.25 |
| 0.3 | 50 | 4 | 8 | 4.2 | 2 | 3.25 | 3.3 | 3.28 | .208 | 10.4 | 20.8 | 34.11 | 3.641 |
| 0.0 | 50 | 2 | 4 | | | | | | | | | | |

$\sum w = 64.75$; $\sum wy = 275.28$; $\sum wx = 151.97$; $\sum wxy = 651.76$; $\sum wx^2 = 358.69$;

$\sum wy^2 = 1186.22$; $\bar{x} = \sum wx / \sum w = 2.3$; $\bar{y} = \sum wy / \sum w = 4.25$;

$b = \frac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 2.03$; Regression line: $y = \bar{y} + b(x - \bar{x})$, when y=5, x=2.67; when y=7,x=3.66; when y=3, x=1.7

Variance = 0.00231; Fiducial limits- $m_1 = 2.4$; $m_2 = 3.0$; $X^2 = 21.51$

**4e Mutoko: Abamectin**

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l. | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+3.9) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 0.0234 | 50 | 40 | 80 | 78.7 | 2.3 | 5.81 | 5.8 | 5.8 | .453 | 22.65 | 52.1 | 131.37 | 5.9 |
| 0.0117 | 50 | 28 | 56 | 53.2 | 2.0 | 5.08 | 5.1 | 5.08 | .546 | 27.3 | 54.6 | 138.68 | 5.1 |
| 0.00585 | 50 | 13 | 26 | 21.3 | 1.7 | 4.19 | 4.2 | 4.2 | .356 | 17.8 | 30.26 | 74.76 | 4.3 |
| 0.0 | 50 | 3 | 6 | | | | | | | | | | |

$\Sigma w = 67.75$; $\Sigma wy = 344.81$; $\Sigma wx = 136.96$; $\Sigma wxy = 706.65$; $\Sigma wx^2 = 280.47$;

$\Sigma wy^2 = 1780.39$; $\bar{x} = \Sigma wx / \Sigma w = 2.0$; $\bar{y} = \Sigma wy / \Sigma w = 5.1$;

$b = \frac{\Sigma wxy - \bar{x}\Sigma wy}{\Sigma wx^2 - \bar{x}\Sigma wx} = 2.6$; Regression line: $y = \bar{y} + b(x - \bar{x})$, when y=5, x=2.0; when y=7,x=2.7;

Variance = 0.0022; Fiducial limits- $m_1 = 1.91$; $m_2 = 2.09$; $X^2 = 22.42$

**4f Mutoko - Malathion**

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+2.2) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 1.89 | 50 | 35 | 70 | 68.1 | 2.5 | 5.47 | 5.5 | 5.47 | .516 | 25.8 | 64.5 | 141.13 | 5.32 |
| 1.26 | 50 | 19 | 38 | 34 | 2.3 | 4.59 | 5.0 | 4.60 | .542 | 27.1 | 62.3 | 124.66 | 4.82 |
| 0.63 | 50 | 12 | 24 | 19.1 | 2 | 4.12 | 4.1 | 4.13 | .320 | 16 | 32.0 | 66.08 | 4.07 |
| 0.0 | 50 | 3 | 6 | | | | | | | | | | |

$\sum w = 68.9$; $\sum wy = 331.87$; $\sum wx = 158.8$; $\sum wxy = 771.56$; $\sum wx^2 = 368.54$;

$\sum wy^2 = 1618.33$; $\bar{x} = \sum wx / \sum w = 2.3$; $\bar{y} = \sum wy / \sum w = 4.82$;

$b = \frac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 2.5$; Regression line: $y = \bar{y} + b(x - \bar{x})$, when y=5, x=2.4; when y=7,x=3.2; when y=3, x=1.6

Variance = 0.00783; Fiducial limits- $m_1 = 2.2$; $m_2 = 2.5$; $X^2 = 1.93$

**4g Mutoko - Amitraz**

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+2.4) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 1.2 | 50 | 35 | 70 | 68.1 | 2.3 | 5.47 | 5.5 | 5.47 | .516 | 25.8 | 59.34 | 141.13 | 5.4 |
| 0.6 | 50 | 17 | 34 | 29.8 | 2.0 | 4.48 | 4.8 | 4.5 | .520 | 26 | 52.0 | 117.0 | 4.8 |
| 0.3 | 50 | 10 | 20 | 14.9 | 1.7 | 3.96 | 4.0 | 3.96 | .283 | 14.15 | 24.06 | 56.03 | 4.2 |
| 0.0 | 50 | 3 | 6 | | | | | | | | | | |

$\Sigma w = 65.95$; $\Sigma wy = 314.16$; $\Sigma wx = 135.4$; $\Sigma wxy = 653.87$; $\Sigma wx^2 = 281.31$;

$\Sigma wy^2 = 1520.36$; $\bar{x} = \Sigma wx / \Sigma w = 2.05$; $\bar{y} = \Sigma wy / \Sigma w = 4.8$;

$b = \frac{\Sigma wxy - \bar{x}\Sigma wy}{\Sigma wx^2 - \bar{x}\Sigma wx} = 1.94$; Regression line: $y = \bar{y} + b(x - \bar{x})$, when y=5, x=2.1; when y=7,x=3.2; when y=3, x=1.1

Variance = 0.0082; Fiducial limits- $m_1 = 1.92$; $m_2 = 2.28$; $X^2 = 6.7$

**4h Mutoko - Dimethoate**

| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Dose or gai/l. | # of test mites | # of mites dead | % response (death) | Corrected % mortality | Log (+2.3) of dose | Empirical probit | Expected probit | Working probit | Weighting coefficient | Weight | | | Calculated values from the regression line. |
| | n | | | | x | | Y | y | | w | wx | wy | $Y^1$ |
| 0.9 | 50 | 38 | 76 | 74.5 | 2.5 | 5.67 | 5.7 | 5.7 | .477 | 23.85 | 59.63 | 135.95 | 5.5 |
| 0.6 | 50 | 26 | 52 | 48.9 | 2.3 | 4.97 | 5.0 | 5.0 | .542 | 27.1 | 62.33 | 135.5 | 5.1 |
| 0.3 | 50 | 7 | 14 | 8.5 | 2.0 | 3.66 | 3.7 | 3.64 | .177 | 8.85 | 17.7 | 32.21 | 4.4 |
| 0.0 | 50 | 3 | 6 | | | | | | | | | | |

$\sum w = 59.8$; $\sum wy = 303.66$; $\sum wx = 139.06$; $\sum wxy = 715.97$; $\sum wx^2 = 327.84$;

$\sum wy^2 = 1569.66$; $\bar{x} = \sum wx / \sum w = 2.3$; $\bar{y} = \sum wy / \sum w = 5.1$;

$b = \frac{\sum wxy - \bar{x}\sum wy}{\sum wx^2 - \bar{x}\sum wx} = 2.2$; Regression line: $y = \bar{y} + b(x - \bar{x})$, when y=5, x=2.3; when y=7,x=3.2; when y=3, x=1.4

Variance = 0.0035; Fiducial limits- $m_1 = 2.2$; $m_2 = 2.4$; $X^2 = 17.62$

**Appendix 5 Effects of acaricides on natural enemies**

**5a-Ladybird beetle**

| Replication | Abamectin | Malathion | Amitraz | Dimethoate | Control |
|---|---|---|---|---|---|
| 1 | 10 | 8 | 9 | 99 | 0 |
| 2 | 9 | 7 | 9 | 8 | 0 |
| 3 | 10 | 6 | 8 | 6 | 0 |
| 4 | 9 | 9 | 10 | 8 | 0 |
| Total dead | 38/40 | 29/40 | 36/40 | 31/40 | 0/40 |
| % mortality | 95% | 72.5% | 90% | 77.5% | 0% |

**5b Aphid wasp Parasitoids**

| Replication | Abamectin | Malathion | Amitraz | Dimethoate | Control |
|---|---|---|---|---|---|
| 1 | 10 | 10 | 9 | 9 | 0 |
| 2 | 10 | 10 | 9 | 10 | 0 |
| 3 | 9 | 10 | 10 | 10 | 0 |
| 4 | 10 | 9 | 10 | 10 | 0 |
| 5 | 10 | 8 | 10 | 10 | 0 |
| Total dead | 49/50 | 47/50 | 48/50 | 49/50 | 0/50 |
| % mortality | 98% | 94% | 96% | 98% | 0% |

**5c Predatory mites**

| Replication | Abamectin | Malathion | Amitraz | Dimethoate | Control |
|---|---|---|---|---|---|
| 1 | 10 | 9 | 10 | 9 | 0 |
| 2 | 10 | 9 | 10 | 10 | 0 |
| 3 | 10 | 10 | 9 | 10 | 0 |
| 4 | 10 | 10 | 10 | 10 | 0 |
| 5 | 10 | 10 | 10 | 10 | 0 |
| Total dead | 50/50 | 48/50 | 49/50 | 49/50 | 0 |
| % mortality | 100% | 96% | 98% | 98% | 0% |

**Appendix 6 Analysis of variance for natural enemies**

**6a Ladybird beetles**

| Source | Degrees of Freedom | Sum of Squares | Mean Square | F-value | Probability |
|---|---|---|---|---|---|
| Replication | 3 | 618.75 | 206.250 | 2.83 | 0.0989 |
| Treatment | 3 | 1118.75 | 372.917 | 5.11 | 0.0245 |
| Error | 9 | 656.25 | 72.917 | | |
| Non-additivity | 1 | 314.96 | 314.960 | 7.38 | 0.0264 |
| Residual | 8 | 341.29 | 42.661 | | |
| Total | 15 | 2393.75 | | | |

Grand Mean = 84.375 Grand Sum =1350.000 Total Count = 16
Coefficient of Variation = 10.12%

**6b Aphid wasp parasitoid**

| Source | Degrees of Freedom | Sum of Squares | Mean Square | F-value | Probability |
|---|---|---|---|---|---|
| Replication | 4 | 70.00 | 17.500 | 1.00 | 0.4449 |
| Treatment | 3 | 40.00 | 13.333 | 0.76 | 0.5368 |
| Error | 12 | 210.00 | 17.500 | | |
| Non-additivity | 1 | 35.00 | 35.000 | 2.20 | 0.1661 |
| Residual | 11 | 175.00 | 15.909 | | |
| Total | 19 | 320.00 | | | |

Grand Mean = 98.000 Grand Sum = 1960.000 Total Count = 20
Coefficient of Variation = 4.27%

**6c Predatory mites**

| Source | Degrees of Freedom | Sum of Squares | Mean Square | F-value | Probability |
|---|---|---|---|---|---|
| Replication | 4 | 30.00 | 7.500 | 0.16 | 0.9556 |
| Treatment | 3 | 55.00 | 18.333 | 0.39 | 0.7651 |
| Error | 12 | 570.00 | 47.500 | | |
| Non-additivity | 1 | 24.55 | 24.545 | 0.50 | |
| Residual | 11 | 545.45 | 49.587 | | |
| Total | 19 | 655.00 | | | |

Grand Mean = 96.500  Grand Sum =1930.000  Total Count =  20
Coefficient of Variation = 7.14%

**Appendix 7 Analysis of variance for site one**

| Source | Degrees of Freedom | Sum of Squares | Mean Square | F-value | Probability |
|---|---|---|---|---|---|
| Replication | 2 | 267572.17 | 133786.083 | 9.42 | 0.0141 |
| Treatment | 3 | 135050.25 | 45016.750 | 3.17 | 0.1065 |
| Error | 6 | 85186.50 | 14197.750 | | |
| Non-additivity | 1 | 35924.54 | 35924.536 | 3.65 | 0.1145 |
| Residual | 5 | 49261.96 | 9852.393 | | |
| Total | 11 | 487808.92 | | | |

Grand Mean = 279.417 Grand Sum=3353.000 Total Count = 12
Coefficient of Variation = 42.64%

**Appendix 7b Analysis of variance for site two**

| Source | Degrees of Freedom | Sum of Squares | Mean Square | F-value | Probability |
|---|---|---|---|---|---|
| Replication | 5 | 705113.83 | 141022.767 | 8.82 | 0.0005 |
| Treatment | 3 | 334006.83 | 111335.611 | 6.96 | 0.0037 |
| Error | 15 | 239883.17 | 15992.211 | | |
| Non-additivity | 1 | 114040.82 | 114040.816 | 12.69 | 0.0031 |
| Residual | 14 | 125842.35 | 8988.739 | | |
| Total | 23 | 1279003.83 | | | |

Grand Mean = 297.917 Grand Sum =7150.000 Total Count = 24
Coefficient of Variation = 42.45%