

**BIOLOGY AND MANAGEMENT OF THE TWO-SPOTTED
SPIDER MITE, *TETRANYCHUS URTICAE* KOCH
(PROSTIGMATA: TETRANYCHIDAE) ON OKRA
[*ABELMOSCHUS ESCULENTUS* (L.) MOENCH]**

By

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(2011-11-117)

THESIS

*Submitted in partial fulfilment of the
requirement for the degree of*

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DEPARTMENT OF AGRICULTURAL ENTOMOLOGY

COLLEGE OF HORTICULTURE

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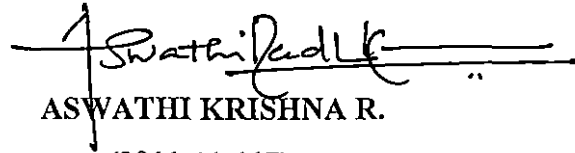
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DECLARATION

I, Aswathi Krishna R. hereby declare that this thesis entitled “Biology and management of the two-spotted spider mite, *Tetranychus urticae* Koch (Prostigmata: Tetranychidae) on okra, [*Abelmoschus esculentus* (L.) Moench]” is a bonafide record of research done by me during the course of research and that the thesis has not previously formed the basis for the award of any degree, diploma, fellowship or other similar title, of any other University or Society.

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CERTIFICATE

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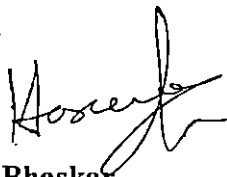
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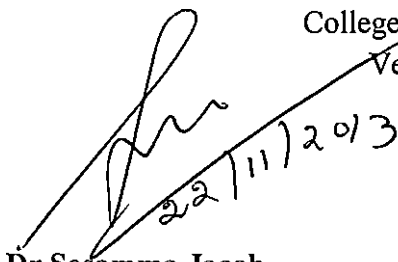
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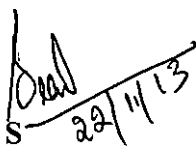
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*This thesis is only a beginning of my
journey...*

*Dedicated to my inspiring parents, brother
and friends for being the pillows, role models,
catapults, cheerleading squad and sounding
boards I have needed.*

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Introduction

1. INTRODUCTION

The sub class Acari which includes mites and ticks, forms an important part of the class Arachnida, with a worldwide distribution and with over 55,000 species described to date (Krantz and Walter, 2009). Among the mites which have successfully colonized the earth, it is the spider mites, belonging to the family Tetranychidae, considered as one of the most studied and observed phytophagous mites. Due to their minute size, these mites remain undetected until major plant damage occurs (Navajas *et al.*, 2010). The damage caused by spider mites to agricultural crops have greatly increased during the past sixty years (Guo-Qing *et al.*, 2010).

The two-spotted spider mite (TSSM), *Tetranychus urticae* (Tetranychidae : Prostigmata) which was first described by Koch in 1836 (Pritchard and Baker, 1955) is an important agricultural pest with a global distribution (Sato *et al.*, 2005). It has been reported to feed on more than 900 host plants and is described as a serious pest of at least 150 economically important agricultural and ornamental plants, including corn, cotton, cucumber, beans, tomato, brinjal, okra and rose (Mondal and Ara, 2006; Kavitha *et al.*, 2007). All stages of TSSM are phytophagous and feed on parenchymatous cells. They typically colonize the undersurface of leaves. Their feeding by desapping result in removal of chlorophyll, yellowing and speckling of leaves, webbing, premature leaf fall, stunting of growth, reduction in photosynthetic activity and transpiration and ultimately death of the whole plant (Damirel and Cabuk, 2008; Varadaraju, 2010). These combinations of effects often reduce the amount of harvestable material.

Okra [*Abelmoschus esculentus* (L.) Moench], belonging to the family Malvaceae, is an economically important vegetable crop in India which is cultivated in an area of 391.8 thousand hectares with a production of 3,974.6 thousand tonnes.

The major okra producing states include West Bengal, Assam, Bihar, Uttar Pradesh, Orissa, Maharashtra, Andhra Pradesh and Karnataka (Anon., 2006). According to Varadaraju (2010), among many factors responsible for low production of okra, the damage inflicted by insect and mite pests has been considered important. About 72 species of pests have been recorded on okra (Rao and Rajendran, 2003). Among these, *T. urticae*, a major pest in summer season, causes 17.46 per cent reduction in yield (Ghosh *et al.*, 1996).

Apart from its polyphagous nature, high reproductive potential and short life cycle, factors such as change in climatic conditions and over-use of plant protection chemicals also have helped to compound the mite problem in okra. Varadaraju (2010) reported that *T. urticae* had developed resistance against several compounds such as organophosphates, dicofol, hexythiazox, clofentezine and fenpyroximate. Though natural enemies play a major role in mitigating the mite population in nature, their population have been greatly declined due to the indiscriminate use of broad spectrum insecticides and acaricides. Increased concern over adverse effects of the commonly used acaricides warrants the development of alternative management strategy including botanicals, acaropathogenic fungi and exploitation of safer acaricide molecules having different modes of action which in turn calls for a thorough understanding regarding the biology of the pest. However, no study on the same has yet been conducted on *T. urticae*, one of the major mite pests of okra in Kerala.

It is in this context that the present study entitled “Biology and management of the two-spotted spider mite, *Tetranychus urticae* Koch (Prostigmata: Tetranychidae) on okra [*Abelmoschus esculentus* (L.) Moench]” was undertaken with the following objectives:

- To study the biology of *T. urticae* on okra.
- To evaluate the bioefficacy of selected acaropathogenic fungi, botanicals and new acaricide molecules against the two-spotted spider mite *T. urticae* on okra.

Review of Literature

2. REVIEW OF LITERATURE

The two-spotted spider mite (TSSM), *Tetranychus urticae* Koch, is an extremely polyphagous pest of numerous vegetables, fruits and ornamentals. This mite causes serious damage and its control is very much essential to get optimum yield of the produce. The intense use of synthetic chemicals against this pest has resulted in the continued development of resistance to a wide range of chemicals. This has necessitated the development of newer chemical groups with novel mode of action. There is also an increasing interest for natural pesticides which are derived from plants and micro organisms since they are perceived to be safer than the synthetic chemicals (Yanar *et al.*, 2011).

2.1. BIOLOGY OF SPIDER MITES

For the successful management of any pest, a thorough knowledge regarding its biology is necessary. The literature pertaining to the biology of *T. urticae* and closely related species are reviewed below. The life cycle of spider mite consists of an egg stage which on hatching passes through three primary stages – the larva, nymph (protonymph and deutonymph) and adult (Krantz, 1970). The immature stages *viz.*, larval and nymphal stages were followed by short quiescent intervals (nymphochrysalis, deutochrysalis and teleiochrysalis), during which moulting takes place (Patil, 2005).

The life span of adult female can be further divided into a short pre-oviposition period, oviposition period lasting for few days and post oviposition period which is longer than the pre-oviposition period (Zhi-Qiang, 2003).

Mallik and Channabasavanna (1983) studied the life history of *Tetranychus ludeni* Zacher at $27 \pm 0.5^{\circ}\text{C}$. The development of egg, larva, protonymph and deutonymph of *T. ludeni* was completed in 106, 32.5, 34.5 and 49 hours

respectively. The total time for development of male and female were recorded as 222 and 200 hours.

Studies conducted by Sudharma (1996) on the biology of *T. ludeni* and *Tetranychus neocaledonicus* Andre on rose, indicated that the developmental period of *T. ludeni* was shorter than *T. neocaledonicus* with a mean total developmental time of 6.86 days for male and 11.7 days for female for former and 7.4 days and 14.2 days for the latter, exhibiting a speedier population build up of *T. ludeni*.

Sathiamma (1991) cited by Abhilash (2001) stated that the females and males of *T. ludeni* completed their egg to adult period in 9.73 days and 9.50 days on an average at 24-30°C with unmated females exhibiting arrhenotokous reproduction.

Adango *et al.* (2006) when studied the development of *T. ludeni* on two leafy vegetable crops viz., amaranth and nightshade, observed that the developmental period of immature stages of *T. ludeni* was shorter on amaranth than on nightshade whereas longevity of female was the same on the two vegetable crops. Total fecundity per female was higher on former. However, no differences were observed in the daily fecundity of *T. ludeni* on the two crops.

The pre-oviposition, oviposition periods and fecundity of mated and unmated females of *T. ludeni* were studied on potted French beans at 19.3 to 28.4°C under field conditions. They were recorded as 1.54 days and 22 days and 165 eggs for mated female. Whereas for unmated females the values were 1.43 days and 27 days and 132 eggs on an average (Puttaswamy and Channabasavanna, 1980).

Studies conducted by Patil (2005) on the biology of *Tetranychus macfarlanei* Baker and Pritchard reported that the total time taken by the mite to complete its life cycle from egg to adult was 10.63 days for male and 12.41 days for female. Egg, larva, protonymph and deutonymph lasted for 3.89, 1.69, 1.86 and 1.23 days in male. Whereas female took 3.89, 2.09, 2.24 and 1.67 days, respectively.

Manjunatha and Puttaswamy (1989) studied the life history of *T. neocaledonicus* on potted French bean plants in the green house at 14.8 to 22.7 °C. Females took 10.44 days and males took 10.19 days to complete their life cycle. The sex ratio was recorded as 1: 10.

Kambrekar and Nandihalli (2003) studied the life history of *T. neocaledonicus* on brinjal and observed that egg, larval, protonymphal and deutonymphal periods ranged from 3.4 to 4 days, 1.5 to 2.5 days, 1 to 2 days and 1 to 1.5 days, respectively. Total life cycle was completed in 7 to 10 days. Studies conducted on the same species on *Moringa oleifera* revealed that *T. neocaledonicus* on an average laid 26.7 ± 0.63 eggs with an oviposition period of 6 days (Kaimal *et al.*, 2007).

Investigations on the fecundity and longevity of mated and unmated females of four phytophagous mites viz., *Tetranychus fijiensis* Hirt, *Tetranychus lambi* Baker and Pritchard, *Tetranychus marianae* McGregor and *T. neocaledonicus* revealed that unmated females of these species lived longer though laid fewer eggs (Bonato and Gutierrez, 1999).

Ying and James (1998) observed that the tumid spider mite, *Tetranychus tumidus* Banks successfully developed from egg to adult at temperatures ranging from 15 to 35°C, but it failed to develop beyond the larval stage at 10°C. However, at 15°C and 35°C, oviposition was greatly reduced to 19.44 and 20.54 eggs, respectively. Similar work done by Bonato (1999) reported that *Tetranychus evansi* Baker and Pritchard took shortest developmental time of 6.3 days at 36°C and maximum fecundity (123.3 eggs) was recorded at 31°C. Vasconcelos *et al.* (2004) also worked out the thermal requirements for *Tetranychus abacae* Baker and Pritchard on *Musa* sp. High fecundity was registered at 25.5°C. It was estimated that this mite could develop up to 31 generations per year at 26°C in field conditions and 31 generations per year at 30°C in green house conditions in Brazil. Similar inverse

relationship between the developmental period and temperature was also reported in strawberry spider mite *Tetranychus turkestanii* Ugarov and Nikolskii and carmine spidermite *Tetranychus cinnabarinus* Boisduval (Northcraft and Watson, 1987; Karami-Jamour and Shishehbor, 2012).

Kazak and Kibritci (2008) reported that the development times of various stages, pre oviposition, oviposition, and post oviposition periods and longevity of *T. cinnabarinus* on eight strawberry cultivars showed no significant host plant effects. But the fecundity of mite was significantly influenced by the strawberry varieties with highest total fecundity of 163.44 eggs and minimum fecundity of 62.71 eggs among the different cultivars.

When *T. ludeni* was reared on velvet bean, *Mucuna deeringiana* in the laboratory at $30 \pm 2^{\circ}\text{C}$ and $70 \pm 5\%$ relative humidity, the development of egg, larva, protonymph and deutonymph was completed in 2.6, 1.06, 1.08 and 1.87 days in males and 3.06, 1.08, 1.10 and 1.89 days in females, respectively. Adult females generally preferred upper surface for egg laying with an average fecundity of 83.6 eggs. Adult longevity was 12.6 and 12 days for males and females (Kaimal and Ramani, 2011).

2.1.1. Biology of *Tetranychus urticae* Koch on different crops

Kasap (2002) studied the biology and life tables of *T. urticae* on three different host plants (bean, cucumber and rose) at $25 \pm 2^{\circ}\text{C}$. Total development time of *T. urticae* was determined as 10.9, 10.4 and 11.2 days on the leaves of bean, cucumber and rose respectively. The sex ratio of *T. urticae* was 0.76, 0.65 and 0.68 on the leaves of bean, cucumber and rose. It was also observed from the study that bean and cucumber were the more suitable hosts compared to rose.

Rajkumar (2003) studied the biology of *T. urticae* on jasmine in the laboratory. He observed that the total time taken by the mite to complete its life

cycle from egg to adult was 10.70 days for male and 12.36 days for female. In male, the egg, larva, protonymph and deutonymph stages lasted for 4.30, 2.42, 1.66 and 1.30 days respectively. Where as in female, 4.46, 2.72, 2.33 and 1.50 days were the periods of developmental stages. Females on an average laid 104 eggs in their oviposition period of 14.5 days. Adult longevity was reported as 12.1 and 18.7 days in males and females.

Sabelis (1981) reported *T. urticae* as an arrhenotokous parthenogenetic species with unmated females producing haploid eggs which develop into males where as mated females produced both haploid and diploid eggs which developed into both males and females.

Silva *et al.* (2009) reported that in gerbera *T. urticae* completed the development of egg, larva, nymphochrysalis, protonymph, deutochrysalis, deutonymph and teleiochrysalis in 3.15, 3.46, 1.13, 2.10, 1.07, 1.63 and 1.11 days respectively. Egg viability was recorded as 96.5 per cent and 97.1 per cent for unmated and mated females.

Life table studies of *T. urticae* under laboratory conditions during summer, autumn and winter reported that the total developmental period of mite from egg to adult varied from 7-8, 9-10 and 17-19 days respectively among the three seasons with a male to female ratio of 1:2.9 (Hoque *et al.*, 2008).

Naher *et al.* (2008) studied the duration of developmental stages of *T. urticae* in different months at room temperature. They observed that during high temperature, the duration of developmental stages were shortened and it took only 4.22 days from egg to adult during the hot months compared to winter period during which the development was completed in 28.33 days.

When El-Wahed and El-Halawany (2012) studied the effect of temperature on the biology of *T. urticae* on pear variety, they observed that with the increase in

temperature the mean incubation period was reduced. The shortest incubation period of 2.7 and 2.6 days at 30°C and the longest duration of 13.6 and 13.4 days at 15°C for female and male were reported. As the temperature increased from 15°C to 30°C, the duration of life stages and life span were also shortened. Rate of reproduction was significantly increased as the temperature increased with highest mean fecundity of 156.8 eggs at 30°C. Riahi *et al.* (2013) also studied the development and reproduction of *T. urticae* on different temperature regime ranging from 13°C to 33°C and found that no development of the mite was observed at 13°C. The developmental time decreased gradually from 17°C to 27°C and increased at higher temperatures (27°C to 33°C). Fecundity recorded at 25, 27, 30 and 33 °C were 40.09, 18.74, 8.03 and 21.33 respectively. Mean longevities of the females were 12.91, 5.92, 3.56 and 6.53 days respectively at the above mentioned temperatures. Two-spotted spider mite can complete a generation as little as 5 days under the most favorable conditions of temperature (Fahnbulleh, 2007).

Razmjou *et al.* (2009) studied the life history traits of *T. urticae* on three legume cultivars in Iran at $25 \pm 1^\circ\text{C}$ and $60 \pm 10\%$ RH. The egg hatchability and development time of the mite were found to be similar among the cultivars with significant variation in fecundity and longevity. Whereas the studies on the biology of *T. urticae* on three pear varieties in Iran at $27 \pm 1^\circ\text{C}$ and $50 \pm 10\%$ relative humidity revealed that though the total mean developmental period of the mite varied among the varieties, the pre-oviposition, oviposition and post-oviposition periods, total fecundity and longevity did not differ significantly among the same (Riahi *et al.*, 2011). When the biology of *T. urticae* was studied on six common bean cultivars at $25 \pm 2^\circ\text{C}$, $70 \pm 5\%$ relative humidity in Iran, significant variation was observed in terms of total developmental period of males and females, oviposition and post oviposition periods, fecundity, longevity and sex ratio among the different cultivars (Najafabadi, 2012).

Tien *et al.* (2011) observed that *T. urticae* females exhibited a choice among the males in order to avoid inbreeding depression as it reduced the offspring fitness. Females preferred to mate with unrelated males. There were significantly higher copulations (64%) with the unrelated males. The inbred off springs matured slower than out bred off springs and inbred adult females had low reproductive output.

2.2. MANAGEMENT OF TETRANYCHID MITES IN VEGETABLES

Tetranychid mites are known as serious pests of several crops cultivated in tropical and subtropical regions. They are one of the important pests of vegetables in India. Short developmental time and high fecundity of these mites coupled with the indiscriminate use of insecticides and varying weather conditions rapidly enable them to develop into a resistant population.

2.2.1. Conventional synthetic acaricides for mite management

Synthetic acaricides are used in mite management since dicofol was discovered during the Second World War. Earlier to this, elemental sulphur was used as an acaricide. Sannaveerappanavar and Channabasavanna (1989) indicated that dicofol is highly effective against all stages of mite when tested against *T. ludeni* in the laboratory. Nine acaricides were evaluated for efficacy against *T. urticae* infesting aubergine in Varanasi, Uttar Pradesh, India during May 2002. Among the acaricides, dicofol 18.5 EC was the most promising against the mite (Siddiqui and Singh, 2006). Mite infestation on jute which was effectively checked with dicofol application has developed resistance against *T. urticae* on tomato in parts of Karnataka. Similarly resistance to dicofol was observed to develop on mite species of okra in parts of Gujarath, necessitating the use of newer acaricidal molecules with novel mode of action (AINPAA, 2009).

After these acaricides, organochlorine compounds like endosulfan having acaricidal properties in addition to their insecticidal activity were used in mite

control. Earlier, Patel (1982) had shown that the use of carbaryl or endosulfan precipitated the outbreaks of *Tetranychus telarius* Linn. in eggplants grown in the field. Use of synthetic pyrethroids has eliminated the natural enemies of mites and at sub-lethal concentrations, stimulated reproduction of mites leading to resurgence of mite pests on crops, mainly vegetables like okra and brinjal (Srinivasa *et al.*, 2010). A similar observation was made by James and Price (2002) with imidacloprid, where resurgence of *T. urticae* by causing an intrinsic increase in egg production in the exposed mites was recorded. To overcome these limitations it is essential that eco-friendly approaches are exploited using botanicals, acaropathogenic fungi and new synthetic acaricides molecules having different modes of action.

2.2.2. Botanicals for the management of spider mites

Probably the most studied botanical insecticide in the last twenty years is a triterpenoid azadirachtin. The major active ingredient of extracts and oils derived from the seeds of *Azadirachta indica*. The effects of azadirachtin is manifested slowly. The studies on spider mites indicate that azadirachtin, in addition to being toxic to various development stages, acts as antifeedant, reduces fecundity and fertility and shortens the life span of adult mites (Marcic *et al.*, 2011).

Srinivasa and Sugeetha (1999) reported that under laboratory conditions, neem oil at two, three and five per cent concentration can cause adult mortality of *T. macfarlanei* to the tune of 22.22, 73.33 and 53.33 per cent respectively.

Laboratory studies on the effect of various plant extracts on *T. urticae* indicated that seed kernel extract of neem (NSKE) has caused 60.25 per cent and 51.2 per cent mortality of the mite at 5 per cent and 1 per cent respectively (Yathiraj and Jagadish, 1999).

Umamahesheswari *et al.* (1999) noticed that among the different neem formulations and castor oil tested for their efficacy against red spider mite, following

dip method in the laboratory, neem oil gave significantly higher mortality (79.60 %) compared to the other treatments tested.

Field experiment on cowpea for managing *T. ludeni* using botanicals proved that neem oil 5 per cent could be successfully used for suppressing the mite population. It was followed by neem garlic 2 per cent. But after five days, the mite population increased which showed the relevance of requirement of repeated application of botanicals (Abhilash, 2001).

Among the various botanicals tested for ovicidal action against *T. macfarlanei*, neem oil proved superiority by recording maximum egg hatch inhibition of 55.18 per cent. Neem oil also caused significantly higher mortality of adults (21.67 %) at 24 hours which was on par with NSKE (18.33 %) (Patil, 2005).

Among the different biorationals tested against tetranychid mites on brinjal, neem oil 2 per cent and NSKE 5 per cent were superior over nimbecidine, azadirachtin, pongamia oil, *Lecanicillium lecanii* (Zimmerman) Gams and Zare and *Metarhizium anisopliae* (Metschnikoff) Sorokin in reducing the mite population. But, they were found inferior to standard check, dicofol. The analysis of yield and net return of various biorationals have indicated that neem oil and NSKE recorded higher yield and maximum net returns (Prasanna, 2007).

Neem oil 2 per cent was found to be the most effective in controlling the mite population on rose cultivated under polyhouse condition which brought out maximum reduction of mite population compared to other botanicals. NSKE 5 per cent was moderately effective in controlling the mite population on rose (Kumar, 2007).

Among the various botanicals studied for efficacy against red spider mite on brinjal in Karnataka, neem oil 2 per cent exhibited maximum acaricidal action (Patil and Nandihalli, 2009). At Varanasi, among the various botanicals tested against

spider mites, NSKE 5 per cent proved to be the best, followed by azadirachtin and mahua oil respectively (AINPAA, 2009).

In the bioassay study on *T. urticae*, neem oil 2 per cent was found to cause a mean mortality of 55 per cent as compared to azadirachtin 0.03 EC, which caused only 16 per cent. This shows that neem oil is promising botanical source for mite management in brinjal (Gauraha and Singh, 2011).

2.2.3. Acaropathogenic fungi for the management of spider mite

The moniliaceous fungus, *Hirsutella thompsonii* Fisher was highly pathogenic to the carmine spider mite, *T. cinnabarinus*. The fungus penetrated the mite's integument mainly through the legs and formed hyphal bodies in chains in the haemolymph. Hyphae on which the spores were produced, began to emerge through the genital and anal apertures and then all over the body. The fungus killed most mites by the second day, at 25°, 27° and 30°C (Gerson *et al.*, 1979). *Beauveria bassiana* (Balsamo) Vuillemin, a filamentous entomopathogenic fungus, belongs to a class of insect pathogenic deuteromycete is also known as imperfect fungus. Strains of *Beauveria* are highly adapted to particular host insects. It causes white muscardine disease by invading directly through the cuticle (Sandhu *et al.*, 2012).

Laboratory bioassay to evaluate the efficacy of *H. thompsonii* was conducted by direct placement of conidia on mites which were placed on bean leaf discs floating on distilled water. A mean mite mortality of 96.5 per cent was recorded (Gardner *et al.*, 1982).

Ovicidal activity of two fungal pathogens, *B. bassiana* and *Paecilomyces fumosoroseus* (Wise) Brown and Smith against *T. cinnabarinus* was studied at 25°C on leaves of *Vicia faba*. The mortality was recorded as 20 per cent and 16 per cent. Though both the fungi were capable of infecting *T. cinnabarinus* eggs, ovicidal activity of *B. bassiana* isolate was higher than that of *P. fumosoroseus*. Egg

mortalities caused by *B. bassiana* infection varied over time but had no change from day 8 or 9 onwards (Shi and Feng, 2004).

Evaluation of formulations of the entomopathogenic fungus *H. thompsonii* against red spider mites on tea showed all the stages of the mite susceptible to the fungus and a maximum mortality of 74 per cent was observed at 12 days post inoculation (UPASI, 2005).

An experiment was carried out to compare direct and indirect application bioassays on the adults of *T. urticae* under laboratory conditions. It was found that *B. bassiana* caused more mortality when indirect application method was used. There was an increase of 26 per cent more mortality in the former (Chandler *et al.*, 2005).

Laboratory bioassay conducted by Wekesa *et al.* (2006) proved that *B. bassiana* was more pathogenic to adults of *T. evansi* compared to *M. anisopliae*. The lethal mortality time of the same varied from 4.6 to 5.8 days.

Aghajanzadeh *et al.* (2006) reported the culture filtrate of *H. thompsonii* to be toxic to the two spotted spider mite recording maximum mortality of 27.97 per cent 8 days after treatment. The mycelia suspension induced highest mortality (57.70 per cent) compared to undiluted filtrate (27.97 per cent).

In bioassay studies conducted on the susceptibility of red spider mite, *T. urticae* against different fungal pathogens like *B. bassiana*, *P. fumosoreases*, *L. lecanii* and *H. thompsonii*, *B. bassiana*, isolated from naturally infected population of *T. urticae* was found to be more virulent against *T. urticae* adult (Ghosh *et al.*, 2007).

A survey conducted in Karnataka during 2004 – 2005 for natural occurrence of fungal pathogen of tetranychid mites on tomato, beans and okra recorded *B. bassiana* isolates with an infection rate of 12.94 per cent on beans (Kalmath *et al.*, 2007).

Studies on the efficacy of different entomopathogenic fungi in combination with adjuvants against *T. urticae* on okra in green house revealed that *H. thompsonii* was the best among the fungi with 75.6 per cent reduction in the mite population and it was on par with abamectin spray (NBAIL, 2007). A rapid decline in the population of *T. urticae* was recorded in green house cultivated bean plants on sixth day after the second spray of *B. bassiana* which was statistically superior over *P. fumosoroseus* (Kalmath *et al.*, 2008).

Twenty-six isolates of entomopathogenic fungi *B. bassiana* and *M. anisopliae* from International Centre of Insect Physiology and Ecology (ICIPE) culture collection, were tested in the laboratory to determine their pathogenicity to adult *T. urticae* and *T. evansi*. All the fungal isolates were pathogenic to the two spider mite species, causing mortality ranging between 95.2 to 99.0 per cent (*B. bassiana*) and 36.5 to 100 per cent (*M. anisopliae*) in *T. urticae* and between 83.0 to 95.2 per cent (*B. bassiana*) and 30.4 to 90.5 (*M. anisopliae*) in *T. evansi*. The lethal time to 50 per cent mortality values varied from 3.0 to 8.3 days with *T. urticae* and from 4.7 to 8.2 days with *T. evansi* Baker & Pritchard (Bugeme, 2008).

Naik and Shekharappa (2009) during *in vitro* evaluation of entomopathogenic fungal formulations against mite pest on okra noticed that the efficacy against mite was 96.67 and 94.67 per cent in *M. anisopliae* oil and wettable powder (WP) formulations followed by *B. bassiana* oil formulation (94 per cent). However, the standard check (Oxydemeton methyl 25 EC) recorded cent per cent mortality throughout the experimental period.

Draganova and Simova (2010) conducted bioassays with different isolates of the entomopathogenic fungus *B. bassiana* under laboratory conditions to estimate their virulence to the two-spotted spider mite *T. urticae*. Mycosis caused to *T. urticae* by the *B. bassiana* isolates 444 Bb and 445 Bb had fast lethal effect after treatment with conidial suspensions even at the concentration of 10^6 conidia ml⁻¹. The mean

mortality values of host individuals were 83.78 ± 3.62 per cent and 68.49 ± 4.28 per cent on the first day and up to 100 per cent in both variants on the fourth day respectively.

Laboratory experiment was conducted on mite-infested excised-leaves of tea in order to test the virulence of locally isolated strain of *H. thompsoni* on tea mite. It was found effective in reducing mite populations giving about 65 per cent mite mortality (Amarasena *et al.*, 2011).

Pathogenicity studies on the fungi *Fusarium semitectum* Berk and Rarenel and *H. thompsonii* by Rachana *et al.* (2012) showed that *H. thompsonii* was more effective than *F. semitectum* in the management of *T. neocaledonicus* causing mortality of immature stages and adults to the tune of 82.05 and 92.31 per cent as compared to *F. semitectum* which recorded 78.20 and 88.34 per cent mortality respectively.

Surveys undertaken during 2009 – 2010 to isolate potential entomopathogenic fungi against spider mites identified *Cladosporium cladosporioides* (Fresen.) de Vries and *B. bassiana* from cow pea, red gram and okra. On assessing the efficacy against *T. urticae* on okra, *C. cladosporioides* was found to be more effective, recording 96.75 per cent mortality as compared to *B. bassiana*, which caused only 5.25 per cent mortality of mites (Jeyarani *et al.*, 2012).

Sayed-Talebi *et al.* (2012a) reported that the mortality of adult female *T. urticae* increased significantly when *B. bassiana* was applied with 5 and 10 per cent concentrations of *Ginkgo biloba* extract showing a synergistic effect between *B. bassiana* and ether-extract of *G. biloba* in controlling the two-spotted spider mite. Similarly the synergistic effect of *B. bassiana* and spiroticlofen on two-spotted spider mite was studied by Sayed-Talebi *et al.* (2012b) and observed that *B. bassiana* alone caused 48.6 per cent mortality while it was 80.6 per cent when *B. bassiana* was

applied along with spiroticlofen. The results demonstrated the synergistic effect of *B. bassiana* and the acaricide spiroticlofen.

Effects of the entomopathogenic fungus *B. bassiana* were studied on life table parameters of two-spotted spider mite, *T. urticae* feeding on bean and cucumber under laboratory conditions. The developmental periods for all immature stages were not affected by fungal infection on each host plant but the duration of larval stage was significantly longer on bean. The female and male longevity, oviposition period and fecundity were significantly lower on fungus treated mites (Seyed-Talebi *et al.*, 2012c).

A formulation of *B. bassiana* called Naturalis was tested for its virulence against *T. urticae* infesting vegetables grown under green house conditions. The mycopesticide Naturalis, applied at 0.1 per cent concentration against *T. urticae* on cucumber reduced mite population density by 85-86 per cent and on tomato it was 93 per cent (Marcic *et al.*, 2012).

2.2.4. New acaricide molecules for the management of spider mites

Among the new molecules which are commercially available or in pipeline are; macrocyclic lactones – abamectin and milbectin, organophosphates – profenophos, organotins – cyhexatin, fenbutain oxide, pyrazoles – fenpyroximate, growth regulators – buprofezin, flufenoxuron, quinazolins – fenazaquin, thiourea compounds – diafenthiuron, sulphur ester – propargite, tetrionic acid derivatives – spiromesifen. Among these, literatures pertaining to the molecules tested in the present study for efficacy against tetranychid mites are reviewed below.

Diafenthiuron is a contact and stomach acting novel insecticide/acaricide belonging to the group thiourea. It acts by inhibiting the mitochondrial respiration. Spiromesifen is a novel contact insecticidal/acaricidal compound derived from spirocyclic tetrionic acids that acts effectively against whiteflies and mites *via*

inhibition of acetyl-CoA-carboxylase, a lipid metabolism enzyme. Fenazaquin is an acaricide which belongs to quinazoline class of chemicals. It has contact and stomach action. Fenazaquin disrupts the biochemistry of insect mitochondria. There are several reports on the efficacy of these molecules against spider mites infesting different crops.

Field trials conducted at South Africa to test the efficacy of newer molecules against *T. cinnabarinus* in cotton indicated that abamectin, diafenthiuron and bifenthrin had sufficient residual activity to control the mite for up to 8 days. When aerial sprays of diafenthiuron were applied according to an economic threshold of 2 mites/leaf, protection was obtained for about 18 days (Brits and Vickers, 1990).

In polyhouse condition, red spider mite population could effectively be reduced by the use of diafenthiuron (Onkarappa and Puttaswamy, 1999). Diafenthiuron and spiromesifen proved significantly effective with more than 96 per cent mortality of adult mites over other treatments. Dicofol and propargite were next best chemicals with 93.33 and 85.00 per cent mortality. As the mortality increased with increase in duration, diafenthiuron spiromesifen and dicofol excelled in their acaricidal activity with cent per cent kill of mites. Fenazaquin 10 EC emerged as the next best treatment with 90.52 per cent mortality (Patil, 2005).

The ovicidal effect of spiromesifen 240 SC on the red spider mite (*T. evansi*) was 100 per cent. The chemical however did not have acute toxicity on the motile stages of the mites, but completely deterred the adult females from laying eggs (Machini, 2005).

In baseline susceptibility studies conducted by Nauen *et al.* (2005), spiromesifen did not have a good effect on *T. urticae* adult females but, it was highly toxic against eggs of this mite. Marcic *et al.* (2009) reported significant decrease in

fecundity and fertility of *T. urticae* females and reduced viability of eggs treated with spiromesifen.

Patil and Nandihalli (2007) reported diafenthiuron, spiromesifen and dicofol as excellent acaricides in many vegetable crops which caused more than 98 per cent egg mortality and were significantly superior to fenazaquin, recording 93.07 per cent egg mortality. The same study against the adults of *T. macfarlanei* on brinjal showed spiromesifen and diafenthiuron as best acaricides with mean mortality of 99.44 and 98.89 per cent.

Patil and Nandihalli (2007) conducted preliminary studies on the ovicidal action of various chemicals on *T. macfarlanei* infesting brinjal. Diafenthiuron 50 WP and spiromesifen 240 SC proved excellent by causing more than 98 per cent egg mortality. They were significantly superior over other treatments and on par with dicofol. Fenazaquin 10 EC emerged as the next best treatment by recording 93 per cent egg mortality.

In the laboratory bioassay conducted by Negi and Gupta (2007) no larva of *T. urticae* was found to survive for more than 48 hours in fenazaquin treated apple leaves. There was also no adult survival in fenazaquin treatment, proving it to be a promising candidate for mite management.

Onkarappa *et al.* (2007) when conducted field trial for the management of *T. urticae* on brinjal observed that spiromesifen 240 SC had resulted in significant reduction in mite and egg population seven days after treatment, which was on par with dicofol 18.5 EC. The next best treatment was fenazaquin 10 EC. On 14 days after spraying, all the three treatments were reported to be on par in reducing the mite population.

Experiments conducted separately at Ludhiana and Karnataka for the management of spider mites occurring on tomato indicated that newer chemicals,

fenazaquin, propargite and abamectin significantly reduced the mite population for a period of two weeks (AINPAA, 2009).

Varadaraju (2010) reported abamectin as significantly superior to diafenthiuron, fenazaquin, propargite, spiromesifen, chlorfenapyr, dicofol and sulphur in decreasing order of efficacy for reducing the population of red spider mite infesting okra.

Field study to evaluate the efficacy of new acaricide molecules against *Tetranychus spp.* in brinjal indicated that spiromesifen 240 SC excelled in acaricidal activity recording the lowest mite population of 5.90 mites per 6.25 cm² leaf area followed by diafenthiuron and dicofol which recorded 6.17 and 6.58 mites per 6.25 cm² leaf area, respectively. propargite and milbemectin proved to be inefficient in controlling the mite population (Sarma, 2010).

Two different dosages of spiromesifen (45 and 60g a.i. ha⁻¹) when compared with a unique dosage of two commercial formulations of abamectin and fenazaquin (60 and 110 g a.i. ha⁻¹), spiromesifen proved very effective in the control of phytophagous mites at both doses. Its effectiveness demonstrated to be remarkable for approximately one month after application. In contrast, abamectin and fenazaquin had proven their effectiveness for a much lower period of time of about the fifteen days post application (Fanigliulo *et al.*, 2010; Kavitha *et al.*, 2006).

In the experiments conducted at Anand Agricultural University for evaluating the bio- efficacy of fenazaquin 10 EC against *T. urticae* on brinjal revealed that tetranychid mites could be effectively controlled by fenazaquin 10 EC @ 2ml l⁻¹ (Patel *et al.*, 2011).

Field trial conducted at Agrahara village near Bangalore to evaluate new acaricide molecules against spider mites infesting okra revealed that fenazaquin 10 EC reduced mite population to the tune of 58 per cent 10 days after spray. Egg count

was recorded minimum (51.4 per cent) at 3 days after the spray application for the same. Diafenthiuron 50 WP offered maximum control up to 7 days after treatment. However both the treatments were superior to dicofol (AINPAA, 2011).

A field trial was conducted at the Central Research Station Farm, Orissa University of Agriculture & Technology, Bhubaneswar to test the bio-efficacy of fenazaquin 10 EC against *T. urticae* which revealed that fenazaquin at 125 and 150g a.i. ha⁻¹ registered significantly lowest mite population followed by dicofol at 250g a.i. ha⁻¹. Plots receiving fenazaquin at 125 and 150g a.i. ha⁻¹ treatments recorded significantly highest fruit yield (Misra, 2011).

At vegetable research farm Ludhiana, in the field trial to evaluate newer molecules, horticultural mineral oil (HMO) and NSKE, fenazaquin 10 EC at 400g a.i. ha⁻¹ recorded 77.2 per cent reduction in egg population, which was on par with dicofol. NSKE 5 per cent was on par with HMO 1 per cent recording 53.84 per cent reduction in the egg counts (AINPAA, 2011).

An experiment was conducted at Bangalore in farmer's field during summer, to evaluate fenazaquin and HMO, in comparison with the conventional acaricide molecule dicofol against spider mites infesting tomato. Reduction in mite population to the tune of 52 to 81 per cent, 24 to 72 per cent and 46 to 85 per cent was recorded respectively. Fenazaquin offered control over the mite to an extended period of two weeks compared to other treatments (AINPAA, 2011).

In strawberry, fenazaquin application was found to cause significantly higher mortality of spider mites three days after the spray. After seven days, the same proved highly effective by maintaining the mite population under control, which extended up to fifteen days of application (AINPAA, 2011).

Among the different developmental stages studied, the egg stage of *T. urticae* was found to be the most sensitive to the chemical spiromesifen. The fecundity of

treated *T. urticae* females was also significantly reduced. Spiromesifen proved highly toxic to *T. urticae* bringing population suppression in ten days (Sato *et al.*, 2011).

Incidence of all stages of *T. urticae* in two months old papaya plants were successfully contained by application of spiromesifen 240 SC @ 0.06 ml l⁻¹ at Tamil Nadu (Arulprakash and Kavino, 2011). Outbreak of red spider mite in banana at Thrissur district of Kerala during 2012 was also effectively managed by the application of spiromesifen 240 SC (0.8 ml l⁻¹) and fenazaquin 10 EC (2 ml l⁻¹) (Bhaskar *et al.*, 2012).

In the Leaf disc bioassay conducted to study the effect of spiromesifen on eggs and gravid females of *T. urticae*, good ovicidal activity was exhibited by spiromesifen but the survival rate, total number of laid eggs per female and egg hatching rate were greatly reduced (Saryazdi *et al.* 2013).

Fenazaquin 10 EC at 125g ai ha⁻¹ was tested for ovicidal activity in the laboratory against *T. urticae* on okra and recorded 81.25 per cent mortality with only 18.75 per cent egg hatchability (Sangeetha and Ramaraju, 2013).

A study was conducted to assess the bioefficacy of newer acaricides against *T. urticae* on tomato near Bangalore. It was observed that among newer acaricides tested, propargite @ 570g a.i. ha⁻¹, fenpyroximate @ 30g a.i. ha⁻¹ and fenazaquin @ 125g a.i. ha⁻¹ were found more effective against eggs as well as active stages of TSSM on tomato for a period of 15 days. Diafenthuron (@ 350g a.i. ha⁻¹) and chlorfenapyr (@ 75g a.i. ha⁻¹) were next in the order of efficacy (AINPAA, 2013).

Field trial conducted to study the efficacy of acaricides viz. propargite (0.057%), fenazaquin (0.0025%) and hexythiazox (0.005%) against Two Spotted Spider Mite (*T. urticae*) on strawberry during 2011 revealed that maximum mortality of mite was caused by fenazaquin treated plants (0.2 mites per leaf) followed by

propargite (0.6 mites per leaf). Maximum population (9.4 mites per leaf) was recorded in hexythiazox treated plants. After seven days of spray, propargite and fenazaquin proved highly effective by recording 0.6 and 1.4 mites per leaf as against 27.4 and 26.7 mites per leaf in pre-count (AINPAA, 2013).

Polyhouse trial at Himachal Pradesh, to assess the efficacy of three acaricides viz. hexythiazox (0.005%), fenazaquin (0.0025%) and propargite (0.057%) and one Horticulture Mineral Oil, Servo 1 per cent against spider mites in beans produced a maximum kill of mites on fenazaquin and propargite sprayed plants compared to all other treatments (AINPAA, 2013).

Fenazaquin 10 EC and spiromesifen 240 SC when tested along with the conventional acaricide wettable sulphur for efficacy against *T. urticae*, fenazaquin recorded the minimum mite population at two, four and seven days after treatment which was on par with sulfur. Though the population of mites two days after treatment was significantly high in spiromesifen compared to other treatments, it was found to be on par with them at four and seven days after treatment (AINPAA, 2013).

In the pot culture experiment conducted at the insectary of TNAU to test the bioefficacy of fenazaquin 10 EC against *T. urticae* on okra, 92.3 per cent reduction in the mite population was recorded three days after spraying. On seven and ten days after spraying the reduction in mite population was 90.5 and 85.3 indicating that fenazaquin 10 EC can offer long term control over the mites. Field trial was also conducted at two locations using different concentrations of fenazaquin 10 EC. The number of live mites decreased as the number of applications increased for fenazaquin at 75, 100, and 150g a.i. ha⁻¹. For fenazaquin at 125g a.i. ha⁻¹ and dicofol

at 250g a.i. ha⁻¹ there were no reduction in the number of live mites due to the number of applications (Sangeetha and Ramaraju, 2013).

Material & Methods

3. MATERIAL AND METHODS

The present study was conducted in the Department of Agricultural Entomology, College of Horticulture, Vellanikkara during February, 2012 – June, 2013. The thrust area of the investigation was to study the biology of two-spotted spider mite, *Tetranychus urticae* Koch and evaluate selected acaropathogenic fungi, botanicals and new acaricide molecules against *T. urticae* on okra.

The methodology and techniques adopted for conducting various experiments based on the objectives set forth in the studies are presented hereunder.

3.1. Biology of *T. urticae* on okra

The studies on the biology of two-spotted spider mite *T. urticae* on okra were conducted in the Acarology laboratory during October- November, 2012 at $30 \pm 3^{\circ}\text{C}$ and $61.5 \pm 7\%$ relative humidity, using the variety *Arka Anamika*.

3.1.1. Identification of *T. urticae*

Twenty gravid females of *T. urticae*, collected from mite infested okra field were released separately on leaf bits of okra placed with their upper surface down on trays containing saturated sponge pad and a layer of blotting paper. The mites were allowed to lay eggs and after seven to ten days, one female and one male were selected randomly from each leaf bit and slides were prepared. The species identity of *T. urticae* was confirmed by using standard taxonomic key (Zhi-Qiang *et al.*, 2002). The characters such as thumb and claw process of pedipalp, tarsal claw and empodium, duplex setae on tarsi I and II and structure of aedeagus were considered for the same (Plate 1). This nucleus culture was used to mass culture *T. urticae* in the laboratory for the subsequent studies.

Plate 1. Taxonomic characters of *T. urticae*



Plate 1a. Thumb-claw process (40X)



Plate 1b. Empodium & claw (40X)



Plate 1c. Duplex setae on leg I (40X)



Plate 1d. Shape of aedeagus (40X)

3.1.2. Mass culturing of *T. urticae*

T. urticae was mass multiplied in the laboratory on okra leaves placed on plastic trays (23 cm x18 cm x5 cm) lined with well moistened synthetic absorbent sponge and a layer of blotting paper (Plate 2). Leaves were placed upside down on the wet blotting paper and gravid females were released. Leaves were changed every three days to avoid poor nutrition for the mite.

3.1.3. Life history of *T. urticae*

The development and life history traits of *T. urticae* were studied following the leaf disc method (Naher *et al.*, 2006). Leaf discs of 4 cm² area were cut from okra leaves and placed upside down on wet cotton bed in Petri plates of 120 mm diameter (Plate 3) that were covered keeping a slight gap to check excessive evaporation. Twenty gravid females were collected from mass culture and transferred to individual leaf discs at the rate of one female per disc for oviposition. After 24 hours, eggs were counted and fifty eggs, selected at random were transferred to leaf discs of 2 cm² area using a moistened camel hair brush (Plate 4).

3.1.3.1. Morphology and developmental duration of immature stages of *T. urticae*

The development of immature stages of the mite was observed with the help of a stereo binocular microscope at 2 hours interval until they reached maturity. The morphology of different life stages of *T. urticae* was studied. The developmental duration of each stage that is, egg, larva, nymphs and quiescent stages were recorded till adult emergence. On emergence, the adult mites were sexed out to work out the developmental duration of different life stages separately for males and females. The values on developmental duration were expressed as mean days \pm Standard Deviation (SD).



Plate 2. Mass culturing of *T. urticae* on okra leaves



Plate 3. Gravid females released for oviposition on leaf discs (4 cm²)

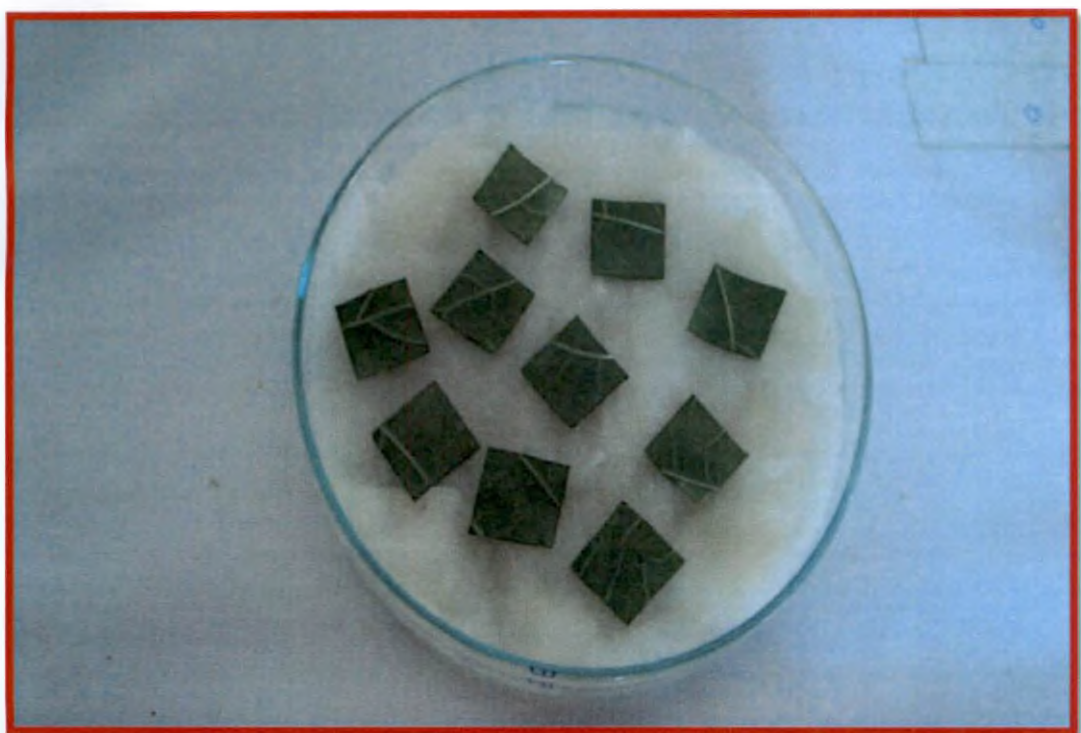


Plate 4. Leaf disc method for studying the biology of *T. urticae*



Plate 5. Mass culturing of *T. urticae* on potted plants of okra in polyhouse

3.1.3.2. Adult longevity

Newly emerged males and females were maintained on separate leaf discs to determine their longevity. Longevity of mated female was determined by placing a newly emerged female on a leaf disc onto which four males were released. The mites were allowed to mate and the males were removed 24 hours later. The mated female was maintained till death. Leaf discs were changed every three days interval. Ten replications each were maintained for males, mated females and unmated females to work out the longevities and expressed in mean days \pm standard Deviation (SD).

3.1.3.3. Reproductive biology of *T. urticae*

To determine the duration of sexual development of mated female, one female teleiochrysalis was transferred to a leaf disc and four adult males were released onto the disc and allowed to mate after the final moult. The males were removed 24 hours after the emergence of the female. The reproductive biology of unmated female was also studied using teleiochrysalis that moulted to female but not allowed to mate. Ten replications each were maintained for mated and unmated females. Observations on mating behaviour, pre-oviposition, oviposition and post-oviposition periods were recorded. The number of eggs laid by the mated as well as unmated females were recorded by replacing the leaf discs carrying eggs with fresh discs till death of the female. The values were expressed as mean number of eggs per female \pm Standard Deviation (SD).

3.1.3.4. Sex ratio and viability of eggs

Sex ratio and viability of eggs were studied following the method described by Gotch and Nagata (2001). The eggs laid by each mated as well as unmated female for the first five days were reared and the viability was determined by counting the number of eggs hatched out to larvae. From this the per cent egg viability was worked out. The emerging mites were sexed out after reaching

adulthood to determine the sex ratio. Ten replications each were maintained for both mated and unmated females.

3.1.3.5. Morphometric parameters of developmental stages of *T. urticae*

Ten individuals of different developmental stages from egg to adult were randomly selected and morphometric parameters were recorded. The diameter of egg and maximum body length and width of other developmental stages were recorded in micrometers (μm) using phase contrast microscope (Leica DM 500) equipped with image analyzer and expressed as mean \pm Standard Deviation (SD).

3.2. Evaluation of selected acaropathogenic fungi, botanicals and new acaricide molecules against the two-spotted spider mite *T. urticae* on okra

3.2.1. Mass culturing of *T. urticae*

T. urticae culture was raised on three weeks old potted plants of okra (variety *Arka Anamika*) in the polyhouse at AINPAA, Department of Agricultural Entomology (Plate 5). The potted plants were periodically replanted to reduce the effect of plant age on mite development and fecundity. New mite colonies were initiated by placing a *T. urticae* colonized okra leaf onto the leaf of a new culture plant.

3.2.2. Field study

Field study was conducted to evaluate the efficacy of two acaropathogenic fungi viz., *Hirsutella thompsonii* Fisher and *Beauveria bassiana* (Balsamo) Vuillemin, two botanicals viz., neem oil 2 per cent and neem seed kernel extract (NSKE 5 per cent) and three new acaricide molecules that is spiromesifen 240 SC, fenazaquin 10 EC and diafenthiuron 50 WP along with a standard check, dicofol 18.5 EC and an untreated control (Table 1) against *T. urticae* on okra, using the variety *Arka Anamika*. The experiments were carried out at College of Horticulture, Vellanikkara during two seasons viz., February – May, 2012 (Season I) and

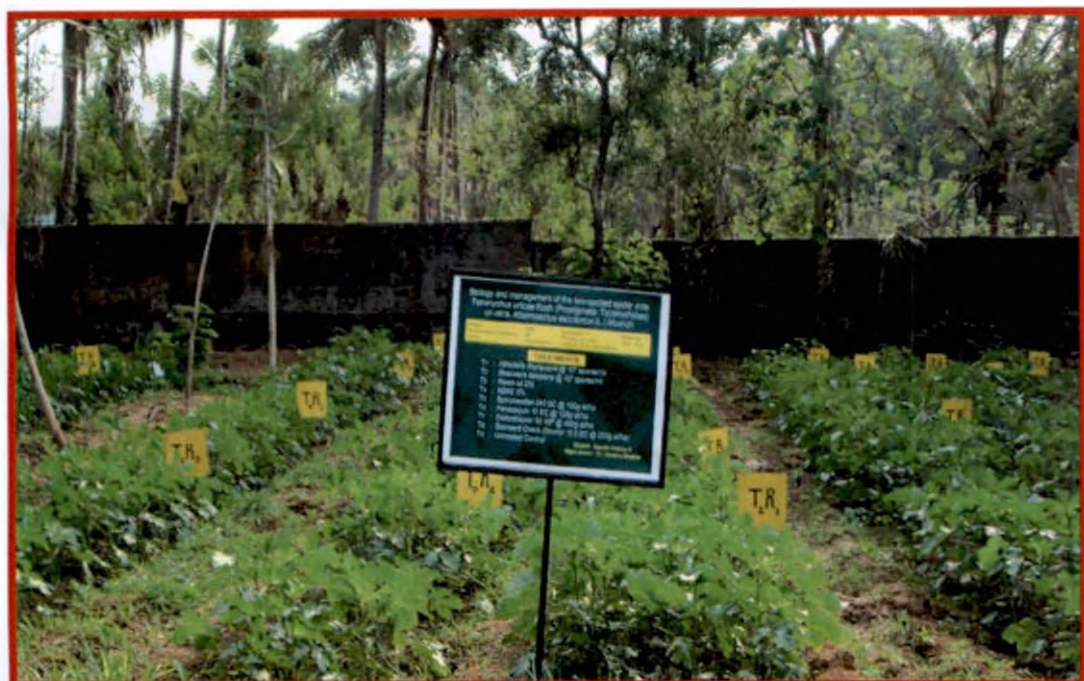


Plate 6. Layout of experimental plot

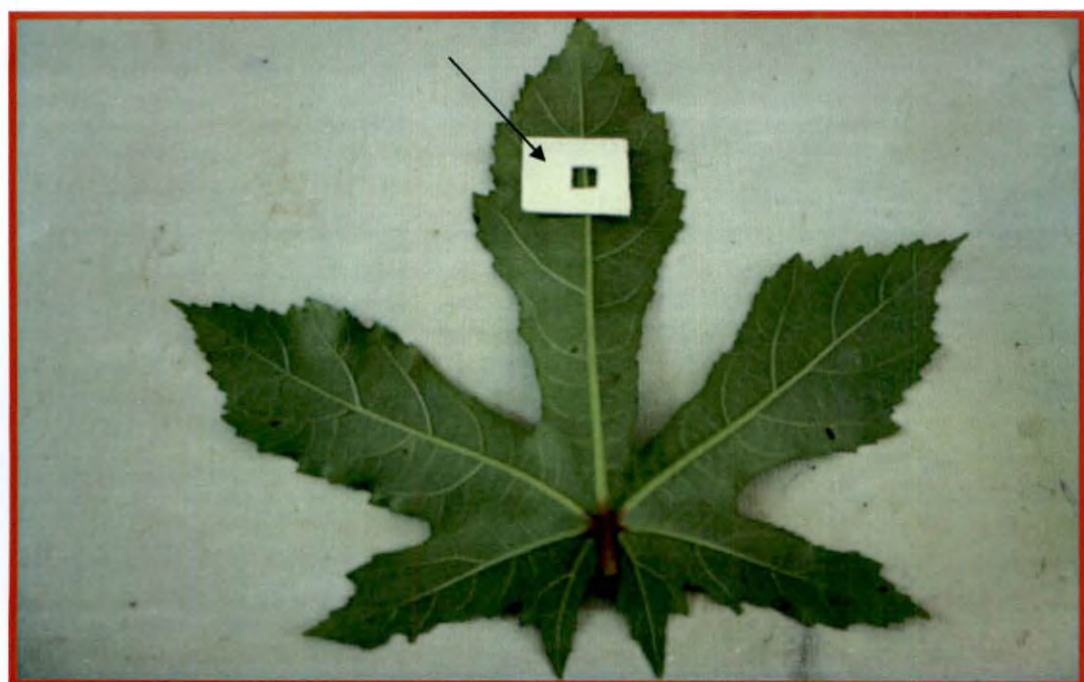


Plate 7. 1 cm² window for counting eggs and mites under microscope

November, 2012 – February, 2013 (Season II). The crop was raised in the field as per the Package of Practices Recommendations (KAU, 2011) at a spacing of 60 x 30 cm in plots of 3 x 3 m size. The experimental layout was Randomized Block Design with three replicates per treatment (Plate 6).

Mites were released on six weeks old plants by stapling mite infested okra leaf bits of 5 cm² size carrying an average of 32 mites per bit at the rate of one bit per plant on the top leaf.

Treatments were imposed three weeks after the release of mites. Spray solution was prepared by thorough mixing of measured quantity of the insecticide and required amount of water to form a uniform emulsion. The treatments were applied using a hand operated high volume knapsack sprayer. Population counts of eggs and active stages of *T. urticae* were recorded from three windows of 1 cm² each from three leaves per plant representing the top, middle and bottom canopy. The population counts were recorded one day before spraying and 3rd, 7th, 10th and 14th day after spraying (Plate 7). The mean mite and egg counts were worked out and analyzed statistically.

3.2.3. Statistical analysis and interpretation of data

Data on mean population of active stages and egg counts were transformed using square root transformation. Population differences on three, seven ten and fourteen days after treatment application were first tested by one way ANOVA. Subsequently the transformed data were analyzed by analysis of covariance (ANOCOVA), taking population counts prior to the first application as covariate and ANOCOVA was done for three, seven ten and fourteen days observations. The result obtained was subjected to DMRT (Duncan's Multiple Range Test). The mean per cent reduction in population over untreated control of mites and eggs was also worked out fourteen days after treatment application.

3.2.4. Meteorological data

Maximum and minimum temperature, relative humidity and rainfall during the cropping period (February 2012 to February 2013) were recorded. The mean weekly values of the meteorological parameters were found out and details are presented in Appendix 1.

Table 1. Treatments evaluated against the two-spotted spider mite *T. urticae* on okra

| Sl. No. | TREATMENTS | Remarks |
|---------|--|---------------------------------|
| 1 | T1: <i>Hirsutella thompsonii</i> @ 10 ⁷ spores/ml | NBAII liquid formulation |
| 2 | T2: <i>Beauveria bassiana</i> @ 10 ⁷ spores/ml | NBAII talc based formulation |
| 3 | T3: Neem oil 2% | Freshly prepared |
| 4 | T4: NSKE 5% | Freshly prepared |
| 5 | T5: Spiromesifen 240 SC @ 100g a.i. ha ⁻¹ | Oberon |
| 6 | T6: Fenazaquin 10 EC @ 125g a.i. ha ⁻¹ | Magister |
| 7 | T7: Diafenthiuron 50 WP @ 400g a.i. ha ⁻¹ | Pegasus |
| 8 | T8: Standard Check (Dicofol 18.5 EC @ 250g a.i. ha ⁻¹) | Hilfol |
| 9 | T9: Untreated Control | - |

Experimental Results

4. EXPERIMENTAL RESULTS

The results of the investigations carried out on the biology and management of the two spotted spider mite, *Tetranychus urticae* are presented in this chapter.

4.1. Biology of *Tetranychus urticae* Koch on okra

The studies on the biology of *T. urticae* were conducted at $30 \pm 3^{\circ}\text{C}$ and $61.5 \pm 7\%$ relative humidity in the Acarology laboratory of Department of Agricultural Entomology, College of Horticulture, Vellanikkara during October- November, 2012.

4.1.1. Life history of *T. urticae* on okra

The life cycle of *T. urticae* consisted of five different stages such as the egg, larva, protonymph, deutonymph and the adult. The larva, protonymph and deutonymph stages were followed by short quiescent intervals called nymphochrysalis, deutochrysalis and teleiochrysalis respectively (Plate 8).

4.1.1.1. Morphology and developmental duration of immature stages of *T. urticae*

The morphological characters and the duration of development of various life stages of *T. urticae* were as follows.

4.1.1.1.1. Egg

T. urticae preferred to colonize and lay eggs on the underside of the leaves of okra. Gravid female mites laid eggs singly or in groups on the webbings as well as on the leaves, often near the veins and the midrib. Eggs were spherical and transparent when freshly laid, but turned creamy white in colour prior to hatching (Plate 8a). During this stage, two dark coloured eye spots, corresponding to the simple eyes of the larvae, were clearly visible. Both males and females of *T. urticae* recorded a mean incubation period of 2.92 days (Table 2).

Table 2. Duration of development stages of *T. urticae* on okra

| Stage | Development period (Mean days \pm SD)* | |
|----------------------------|--|------------------|
| | Male | Female |
| Egg | 2.92 \pm 0.003 | 2.92 \pm 0.003 |
| Larva | 0.83 \pm 0.03 | 1.19 \pm 0.25 |
| Nymphochrysalis | 0.26 \pm 0.01 | 0.68 \pm 0.04 |
| Protonymph | 0.36 \pm 0.03 | 0.58 \pm 0.05 |
| Deutochrysalis | 0.67 \pm 0.03 | 0.29 \pm 0.04 |
| Deutonymph | 0.93 \pm 0.04 | 1.05 \pm 0.05 |
| Teleiochrysalis | 0.76 \pm 0.03 | 0.81 \pm 0.06 |
| Total developmental period | 6.73 \pm 0.18 | 7.52 \pm 0.50 |

*Mean value of fifty observations

4.1.1.1.2. Larva

The eggs hatched out to hexapod larvae. Newly hatched larva was cream coloured and small in size (Plate 8b). On feeding the colour changed from cream to pale green. The simple eyes on the dorso-lateral idiosoma were clearly distinguishable at this stage. The mean larval period recorded was 0.83 days for males and 1.19 days for females (Table 2).

4.1.1.1.3. Nymphochrysalis

The larva stopped feeding and moved to a suitable place on the leaf and entered into its first quiescent stage called nymphochrysalis. During this stage, the anterior two pairs of legs were extended straight forward and kept close to each other. The posterior legs were extended backwards and held close to the sides of opisthosoma (Plate 8c). At the end of this stage moulting took place. Average nymphochrysalis period was 0.26 days for male and 0.68 days for females as presented in Table 2.

4.1.1.1.4. Protonymph

The first nymphal stage the protonymph emerged by splitting open the larval skin along the dorsal midline and was characterized by the presence of four pairs of legs (Plate 8d). Protonymph was larger in size and darker in colour as compared to the larva. The mean protonymph period lasted for 0.36 days for males and 0.58 days for females, as can be seen from Table 2.

4.1.1.1.5. Deutochrysalis

At the end of protonymphal period, the protonymph entered its second quiescent stage, the deutochrysalis. It remained anchored to the leaf surface in a manner similar to nymphochrysalis (Plate 8e). This stage, on an average, lasted for 0.67 days in the case of males and 0.29 days in the case of females (Table 2).

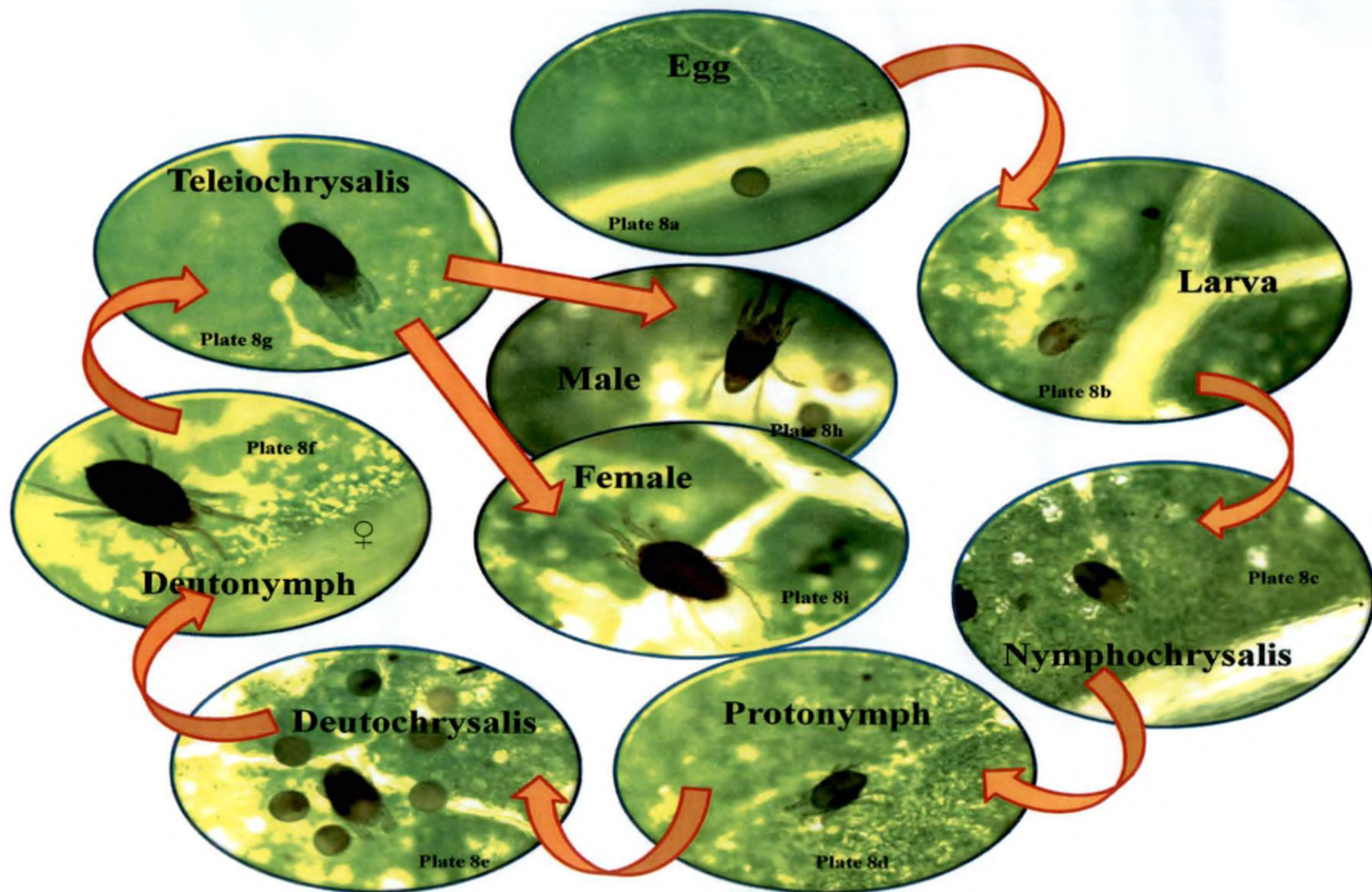


Plate 8. Life cycle of *Tetranychus urticae* on okra

4.1.1.1.6. Deutonymph

Deutochrysalis moulted to the second nymphal stage, the deutonymph. Sexual characters could be distinguished from this stage onwards. Deutonymph was light reddish in colour (Plate 8f). Female deutonymphs were larger and broader than their male counterparts while male deutonymphs were elongate. They were actively moving and feeding. The mean deutonymph period was 0.93 days in for males and 1.05 days for females as presented in Table 2.

4.1.1.1.7. Teleiochrysalis

The deutonymph entered the third quiescent stage, the teleiochrysalis (Plate 9g). This period on an average was 0.76 days for males and 0.81 days for females (Table 2).

4.1.1.1.8. Adult

After the final moult from teleiochrysalis, adult mites emerged. The mites exhibited sexual dimorphism in the adult stage. Males (Plate 8h) were reddish green or light red in colour and smaller in size with body tapering posteriorly to a blunt point. Female mites (Plate 8i) were carmine red in colour, larger and plumper with longer setae over the body and legs. They turned darker after mating. Both males and females had bright red eye spots on the dorso-lateral idiosoma.

4.1.1.1.9. Total development period

The total development period from egg to adult emergence was 6.73 days for males and 7.52 days for females (Table 2).

4.1.1.2. Adult longevity

Adult males recorded a mean longevity of 12 days while the corresponding figures for mated and unmated females were 12.5 days and 17 days respectively (Table 3).

4.1.1.3. Reproductive biology of *T. urticae*

4.1.1.3.1. Mating behaviour

Newly emerged males were found actively moving in search of females to mate. They rested near or over the quiescent female deutonymphs that they came across, guarding (Plate 9) them and even fighting with the rival males (Plate 10) for the yet to emerge females, when a couple or more males were present. Mating took place immediately after the emergence of the female. Male pushed and raised the posterior abdominal region of the female and slide underneath with its hysterosoma upturned. Mating lasted for a mean period of 1.78 minutes (Table 4). Male was observed to mate with several females, though a female usually mated only once. However, females which were surrounded by several males were observed occasionally being attempted to mate by these males if the former stood motionless or were slow moving. But it lasted only for few seconds.

4.1.1.3.2. Pre-oviposition, oviposition and post-oviposition periods

The life span of adult female mites consisted of pre-oviposition period, oviposition period and post-oviposition period, which were observed to be of longer duration in unmated females. The mean pre-oviposition period in mated and unmated females lasted for 0.58 days and 0.84 days respectively. Oviposition and post-oviposition periods lasted for 9 days and 4 days in case of mated females and 11 days and 4.5 days in case of unmated females as represented in Table 4.



Plate 9. Male resting over female teleiochrysalis



Plate 10. Males fighting for female teleiochrysalis

Table 3. Adult longevity of *T. urticae*

| Sex | | Duration (Days \pm SD)* |
|--------|---------|---------------------------|
| Male | | 12 \pm 1 |
| Female | Mated | 12.5 \pm 0.71 |
| | Unmated | 17 \pm 1 |

*Mean of ten observations

Table 4. Pre-oviposition, oviposition and post-oviposition periods

| Parameters | | Duration (Mean \pm SD)* |
|-------------------------|----------------|---------------------------|
| Mating period | | 1.78 \pm 0.36 minutes |
| Pre-oviposition period | Mated female | 0.58 \pm 0.06 days |
| | Unmated female | 0.84 \pm 0.04 days |
| Oviposition period | Mated female | 9 \pm 1.83 days |
| | Unmated female | 11 \pm 1 days |
| Post-oviposition period | Mated female | 4 \pm 1 days |
| | Unmated female | 4.5 \pm 0.71 days |

*Mean of ten observations

4.1.1.3.3. Fecundity, sex ratio and egg viability of *T. urticae*

Mated females on an average laid 108 eggs whereas unmated females laid only 77 eggs. Mated female produced a progeny consisting of both males and females in the ratio 1:5.8 whereas unmated females produced only males. The viability of eggs of *T. urticae* was 92.10 per cent (Table 5).

4.1.2. Morphometric parameters of development stages of *T. urticae*

The morphometrics of different stages are represented in Table 6. They showed a gradual increase in size from egg to adult stage except in the case of teleiochrysalis which shrunk in size as compared to its deutonymph. As sex could be distinguished only after the mites reached their deutonymph stage, the measurements up to this stage were common. Eggs on an average measured 128.59 μm in diameter. The total mean body length and maximum mean body width of larva were 177.13 μm and 122.03 μm . Nymphochrysalis measured 219.51 μm in mean length and 128.65 μm in mean width. The length of the protonymph was 260.61 μ while the width on an average was recorded as 141.99 μm . The deutochrysalis measured 286.86 μm in length and 168.10 μm in breadth. Male deutonymphs had a length of 355.98 μm and a width of 163.51 μm where as female deutonymphs on an average measured 408.20 μm in length and 206.24 μm in width. Length and width of teleiochrysalis were 336.59 μm and 171.32 μm for males and 405.41 μm and 198.63 μm for females. The maximum length and width were recorded during the adult stage for both males and females. It was 345.19 μm and 159.96 μm for males and 488.73 μm and 237.91 μm for females.

Table 5. Fecundity, sex ratio and egg viability of *T. urticae*

| | Fecundity (No. of eggs) | Male : Female ratio | Egg viability (%) |
|------------------|-------------------------|---------------------|-------------------|
| Mated female * | 108 \pm 7.0 | 1: 5.8 | 92.10 \pm 6.85 |
| Unmated female * | 77 \pm 6.56 | 1: 0 | |

*Mean of ten observations

Table 6. Morphometric parameters of development stages of *T. urticae*

| Stage * | Mean \pm SD (μm) | | | |
|-----------------|---------------------------------|-------------------------|--------------------------|-------------------------|
| | Length (μm) | | Width (μm) | |
| Egg (Diameter) | 128.59 \pm 5.77 | | | |
| Larva | 177.13 \pm 8.29 | | 122.03 \pm 10.85 | |
| Nymphochrysalis | 219.51 \pm 8.58 | | 128.65 \pm 3.56 | |
| Protonymph | 260.61 \pm 32.30 | | 141.99 \pm 7.32 | |
| Deutochrysalis | 286.86 \pm 3.39 | | 168.10 \pm 0.51 | |
| | Male | | Female | |
| | Length (μm) | Width (μm) | Length (μm) | Width (μm) |
| Deutonymph | 355.98 \pm 8.45 | 163.51 \pm 4.96 | 408.20 \pm 15.71 | 206.24 \pm 2.74 |
| Teleiochrysalis | 336.59 \pm 2.44 | 171.32 \pm 1.90 | 405.41 \pm 6.22 | 198.63 \pm 1.12 |
| Adults | 345.19 \pm 8.49 | 159.96 \pm 10.21 | 488.73 \pm 12.06 | 237.91 \pm 1.00 |

*Mean of ten observations

4.2. Evaluation of selected acaropathogenic fungi, botanicals and new acaricide molecules against *T. urticae* on okra

A field study was conducted to evaluate the efficacy of two acaropathogenic fungi, two botanicals and three new acaricide molecules along with a standard check and untreated control against the two-spotted spider mite on okra in the experimental plot at College of Horticulture, Vellanikkara during two seasons viz., February – May, 2012 and November, 2012 – February, 2013. Efficacy of various treatments against active stages and eggs of *T. urticae* are presented below.

4.2.2. Efficacy of treatments against eggs of *T. urticae* on okra

4.2.2.1. Season I (February – May, 2012)

The mean egg count of *T. urticae* before the application of treatments ranged from 56.63 to 122.26 per cm² leaf area. The results of the field experiment to evaluate the efficacy of different treatments against eggs of *T. urticae* are presented in Table 7.

At three days after spraying, the new acaricide molecules recorded lower egg counts per cm² leaf area as compared to other treatments. The lowest egg count was recorded by fenazaquin 10 EC (1.96/cm² leaf area) which was also on par with spiromesifen 240 SC (5.85/cm² leaf area) and diafenthiuron 50 WP (5.89/cm² leaf area). These treatments were superior over the standard check, dicofol (28.70/cm² leaf area). Botanicals, neem oil 2 per cent and NSKE 5 per cent recorded 31.93 and 68.22 eggs and were on par with each other. Among the acaropathogens, *Beauveria bassiana* recorded a lower mean egg count of 33.22 as against 80.33 in case of *Hirsutella thompsonii*. However the botanicals and acaropathogens were also on par with the untreated control.

At seven days after treatment, the egg count was found to be low in new acaricide molecules and standard check, dicofol 18.5 EC. Diafenthiuron 50WP

Table 7. Efficacy of various treatments against eggs of *T. urticae* on okra during season I

| TREATMENTS | Mean no. of eggs / cm ² leaf area | | | | | Mean reduction (%) |
|--|--|-------------------------------|-------------------------------|-------------------------------|-------------------------------|--------------------|
| | 1 DBS † | 3 DAS †† | 7 DAS †† | 10 DAS †† | 14 DAS †† | |
| <i>Hirsutella thompsonii</i> @ 10 ⁷ spores/ml | 84.00 (9.10) ^a | 80.33 (8.75) ^a | 22.89 (4.83) ^{ab} | 14.48 (3.83) ^{bc} | 10.94 (3.38) ^{cd} | 25.80 |
| <i>Beauveria bassiana</i> @ 10 ⁷ spores/ml | 63.26 (7.94) ^a | 33.22 (5.58) ^b | 31.74 (5.52) ^a | 26.56 (5.20) ^{ab} | 5.19 (4.99) ^{bc} | 44.22 |
| Neem oil 2% | 106.59 (9.93) ^a | 31.93 (5.74) ^{ab} | 35.70 (6.02) ^a | 53.63 (7.30) ^a | 7.31 (6.21) ^{ab} | 25.84 |
| NSKE 5% | 85.81 (9.24) ^a | 68.22 (8.17) ^{ab} | 38.26 (6.22) ^a | 56.52 (7.40) ^a | 7.40 (4.25) ^{bc} | 1.71 |
| Spiromesifen 240 SC @ 100g a.i. ha ⁻¹ | 56.63 (7.46) ^a | 5.85 (2.20) ^c | 10.15 (3.19) ^{bc} | 21.52 (4.23) ^{bc} | 4.24 (3.54) ^{cd} | 75.91 |
| Fenazaquin 10 EC @ 125g a.i. ha ⁻¹ | 77.18 (8.71) ^a | 1.96 (1.46) ^c | 7.67 (2.73) ^c | 7.55 (2.76) ^c | 2.76 (2.82) ^d | 88.50 |
| Diafenthiuron 50 WP @ 400g a.i. ha ⁻¹ | 74.11 (8.43) ^a | 5.89 (2.40) ^c | 3.89 (2.07) ^c | 9.82 (3.03) ^{bc} | 3.03 (2.49) ^d | 86.95 |
| Standard Check (Dicofol 18.5 EC @ 250g a.i. ha ⁻¹) | 89.82 (9.46) ^a | 28.70 (5.44) ^b | 9.92 (3.19) ^{bc} | 11.93 (3.49) ^{bc} | 3.49 (4.76) ^{bc} | 68.83 |
| Untreated Control | 122.26 (10.46) ^a | 67.48 (8.34) ^{ab} | 43.77 (6.58) ^a | 54.70 (7.44) ^a | 7.42 (6.87) ^a | - |

DBT = Day Before Treatment; DAT = Days After Treatment ; Means followed by same letters do not differ significantly by DMRT ($p = 0.05$) ; Mean reduction- Mean reduction over untreated control; †- Values in the parenthesis are square root transformed values, ††- Values in the parenthesis are adjusted means of square root transformed values based on ANOCOVA

recorded the lowest egg count of 3.89 per cm² leaf area followed by fenazaquin 10 EC (7.67) and spiromesifen 240 SC (10.15). These molecules were on par with each other and standard check dicofol (9.92). The mean egg count of the botanicals were 35.74 in case of neem oil 2 per cent and 38.26 in case of NSKE 5 per cent whereas a mean count of 22.9 eggs per cm² leaf area was recorded in case of *H. thompsonii* as against 31.74 in case of *B. bassiana*. However the botanicals and acaropathogens were on par with each other and untreated control (43.77/cm² leaf area).

Similar trend was observed on ten days after spraying where the chemical acaricides continued to record lower mean egg counts with fenazaquin 10 EC recording the lowest egg count of 7.55 per cm² leaf area. It was also found on par with diafenthiuron 50 WP (9.82), spiromesifen 240 SC (21.52) and standard check (11.93). The botanicals neem oil 2 per cent and NSKE 5 per cent which recorded mean egg counts of 53.63 and 56.52 per cm² leaf area respectively were on par with each other and untreated control (54.70). *H. thompsonii* (14.48) was found to be on par with *B. bassiana* (26.56) but *B. bassiana* was also on par with the botanicals and untreated control.

At fourteen days after spraying, the new acaricide molecules recorded lower egg counts and were superior over other treatments. Lowest egg count was recorded by Fenazaquin (2.76/cm² leaf area) followed by diafenthiuron 50 WP (3.03/cm² leaf area) and spiromesifen 240 SC (4.24/cm² leaf area). All the three new molecules were on par with each other and superior over dicofol 18.5 EC (3.49). Among the botanicals, neem oil 2 per cent recorded a mean egg population of 7.31 per cm² leaf area and was on par with NSKE 5 per cent (7.40). *H. thompsonii* which recorded an egg count of 10.94 per cm² leaf area was again on par with *B. bassiana* (5.19) and superior to untreated control (7.42).

Fenazaquin 10 EC recorded the highest reduction in egg counts of 88.50 per cent which was followed by diafenthiuron 10 EC (86.95%). The next best treatments

were spiromesifen 240 SC and dicofol 18.5 EC with mean per cent reduction in egg count of 75.91 and 68.83. Among the acaropathogenic fungi and botanicals, highest reduction in egg count was exhibited by *B. bassiana* (44.22%) followed by neem oil 2 per cent (25.84%) and *H. thompsonii* (25.80). The botanical NSKE 5 per cent showed only 1.71 per cent reduction in egg count fourteen days after the spray.

4.2.2.2. Season II (November – February, 2013)

The egg counts of *T. urticae* before imposing the treatments ranged from 117.66 to 233.07 per cm² leaf area. The results of the field experiment to evaluate the efficacy of different treatments against eggs of *T. urticae* are presented in Table 8.

At three days after spraying all the treatments except NSKE 5 per cent and *H. thompsonii* significantly reduced the egg count per cm² leaf area compared to untreated control. The lowest egg count of 3.77 per cm² leaf area was recorded by spiromesifen 240 SC followed by fenazaquin 10 EC (5.15) which was on par with diafenthiuron (15.85) and dicofol 18.5 EC (11.70). Among the botanicals, neem oil 2 per cent (59.15) recorded lower egg counts compared to NSKE 5 per cent (93.26). Among the acaropathogenic fungi, *B. bassiana* (68.22) recorded lower egg count per cm² leaf area as compared to *H. thompsonii* (99.04). However both the botanicals and *B. bassiana* were on par with the untreated control (65.67). NSKE 5 per cent and *H. thompsonii* were found to be inferior to all other treatments.

At seven days after spraying, new molecules fenazaquin 10 EC diafenthiuron 50 WP and dicofol 18.5 EC continued to record lower egg per counts per cm² leaf area compared all other treatments. Among them, the lowest mean egg population was recorded by fenazaquin 10 EC (0.77), followed by diafenthiuron 50 WP (2.07) and spiromesifen 240 SC (3.70). However all the four chemicals were on

Table 8. Efficacy of various treatments against eggs of *Tetranychus urticae* on okra during season II

| TREATMENTS | Mean egg counts/ cm ² leaf area | | | | | Mean reduction (%) |
|--|--|-------------------------------|-------------------------------|-------------------------------|------------------------------|--------------------|
| | 1DBS† | 3DAS†† | 7DAS†† | 10DAS†† | 14DAS†† | |
| <i>Hirsutella thompsonii</i> @ 10 ⁷ spores/ml | 200.67 (13.986) ^a | 99.04 (9.88) ^a | 28.00 (5.42) ^a | 19.00 (4.31) ^{ab} | 2.85 (1.69) ^b | 2.79 |
| <i>Beauveria bassiana</i> @ 10 ⁷ spores/ml | 233.07 (15.28) ^a | 68.22 (8.09) ^{bc} | 21.89 (4.85) ^{ab} | 7.11 (2.86) ^{bc} | 3.96 (1.94) ^b | 33.94 |
| Neem oil 2% | 169.08 (12.98) ^a | 59.15 (7.68) ^c | 11.29 (3.37) ^{bc} | 4.48 (2.20) ^{cd} | 0.96 (1.14) ^b | 50.46 |
| NSKE 5% | 174.52 (13.13) ^a | 93.26 (9.65) ^{ab} | 32.52 (5.66) ^a | 11.04 (3.24) ^{bc} | 6.07 (2.44) ^b | 6.71 |
| Spiromesifen 240 SC @ 100g a.i. ha ⁻¹ | 159.30 (12.53) ^a | 3.77 (2.09) ^e | 3.70 (1.97) ^{cd} | 3.07 (1.85) ^{cd} | 2.15 (1.63) ^b | 91.71 |
| Fenazaquin 10 EC @ 125g a.i. ha ⁻¹ | 160.27 (12.54) ^a | 5.15 (2.36) ^{de} | 0.77 (1.06) ^d | 0.33 (0.88) ^d | 0.82 (1.09) ^b | 95.38 |
| Diafenthiuron 50 WP @ 400g a.i. ha ⁻¹ | 170.00 (12.99) ^a | 15.85 (4.01) ^d | 2.07 (1.59) ^{cd} | 1.18 (1.23) ^d | 2.67 (1.78) ^b | 85.79 |
| Standard Check (Dicofol 18.5 EC @ 250g a.i. ha ⁻¹) | 117.66 (10.56) ^a | 11.70 (3.47) ^{de} | 4.66 (1.89) ^{cd} | 0.93 (1.02) ^d | 0.77 (1.12) ^b | 88.21 |
| Untreated Control | 167.00 (12.92) ^a | 65.67 (8.13) ^{bc} | 32.59 (5.71) ^a | 29.96 (5.48) ^a | 24.94 (4.79) ^a | - |

DBT = Day Before Treatment; DAT = Days After Treatment ; Means followed by same letters do not differ significantly by DMRT ($p = 0.05$) ; Mean reduction- Mean reduction over untreated control; †- Values in the parenthesis are square root transformed values, ††- Values in the parenthesis are adjusted means of square root transformed values based on ANOCOVA

par with each other and standard check, dicofol 18.5 EC (4.66). Neem oil 2 per cent, which recorded a population of 11.29 per cm² leaf area was superior to NSKE 5 per cent (32.52) and untreated control (32.59). The fungal pathogens *B. bassiana* and *H. thompsonii* recorded mean egg counts of 21.89 and 28.00 per cm² leaf area and were on par with each other and untreated control.

At ten days after spraying, all the treatments except *H. thompsonii* significantly reduced the egg counts per cm² leaf area as compared to untreated control. Lowest egg count was recorded by fenazaquin 10 EC (0.33), which was on par with diafenthiuron 50 WP (1.18), spiromesifen 240 SC (3.07) and dicofol 18.5 EC (0.93). Neem oil 2 per cent recorded a mean egg count of 4.48 as against 11.04 per cm² leaf area in case of NSKE 5 per cent. However the botanicals were on par with each other, but superior over untreated control (29.96). Among the fungal pathogens *B. bassiana* which recorded an egg count of 7.11 per cm² leaf area was on par with *H. thompsonii* (19.00) and the botanicals. But *H. thompsonii* was also on par with the untreated control.

All the treatments except untreated control were found to be on par with each other fourteen days after spraying and were also superior over untreated control with respect to the mean egg counts. The lowest egg population of 0.77 was recorded by dicofol 18.5 EC followed by fenazaquin 10 EC (0.82). Diafenthiuron 50 WP and spiromesifen 240SC recorded 2.67 and 2.15 egg counts respectively. The mean egg counts of botanicals neem oil 2 per cent and NSKE 5 per cent were 0.96 and 6.07 where as that of *H. thompsonii* and *B. bassiana* were 2.85 and 3.96 per cm² leaf area.

Fenazaquin 10 EC recorded the highest per cent reduction in egg count (95.38) followed by spiromesifen 240 SC (91.71 %). The next best treatments were dicofol 18.5 EC (88.21%) and diafenthiuron 50 WP (85.79%). Among the botanicals, neem oil 2 per cent recorded the highest per cent reduction of 50.46 as

compared to NSKE 5 per cent (6.71%) and among the acaropathogens *B. bassiana* (33.94) exhibited higher mean reduction in egg count than *H. thompsonii* (2.79%)

4.2.1. Efficacy of treatments against active stages of *T. urticae* on okra

4.4.1.1. Season I (February – May, 2012)

The results of the field experiment to evaluate the efficacy of different treatments against active stages of *T. urticae* are presented in Table 9. The mean population of active stages of mite one day before imposing the treatments ranged from 21.96 to 68.48 per cm² leaf area. All the treatments except *H. thompsonii* significantly reduced the population of active stages of mite three days after spraying, as compared to untreated control. The mean mite population ranged from 1.63 to 51.18 per cm² leaf area. All the three new acaricide molecules viz., fenazaquin 10 EC, spiromesifen 240 SC and diafenthiuron 50 WP recorded significantly lower mite populations compared to other treatments. Among the new molecules, fenazaquin 10 EC recorded the lowest population of 1.63 mites per cm² leaf area which was on par with spiromesifen 240 SC (2.67), diafenthiuron 50 WP (3.52) and standard check dicofol which recorded a mean population of 2.55 per cm² leaf area. Among the two botanicals tested, neem oil 2 per cent recorded a lower mean population of 12.33 as against 28.04 mites per cm² leaf area in case of NSKE 5 per cent. However both the botanicals were on par with each other. The mite population was found to be significantly lower in *B. bassiana* treated plots (18.59) as against 39.44 per in *H. thompsonii* treated plots and was on par with the botanicals.

At seven days after treatment, only the new acaricide molecules and dicofol 18.5 EC significantly reduced the mite population as compared to untreated control (21.30). The lowest mite population of 1.96 was recorded by fenazaquin 10 EC followed by diafenthiuron 50 WP (2.55), standard check dicofol (2.67) and spiromesifen 240 SC (3.33). There was no significant difference between any of the chemicals. In the plots treated with botanicals, lower mite population was recorded

Table 9. Efficacy of treatments against active stages of *T. urticae* on okra during season I

| TREATMENTS | Mean no. of mites / cm ² leaf area | | | | | Mean reduction (%) |
|--|---|-------------------------------|-------------------------------|-------------------------------|------------------------------|--------------------|
| | 1 DBT † | 3 DAT †† | 7 DAT †† | 10 DAT †† | 14 DAT †† | |
| <i>Hirsutella thompsonii</i> @ 10 ⁷ spores ml ⁻¹ | 35.11 (5.92) ^a | 39.44 (6.32) ^{ab} | 36.56 (6.01) ^a | 17.28 (4.21) ^{ab} | 11.89 (3.50) ^a | 43.70 |
| <i>Beauveria bassiana</i> @ 10 ⁷ spores ml ⁻¹ | 34.18 (5.76) ^a | 18.59 (4.28) ^c | 16.89 (4.12) ^b | 30.63 (5.07) ^{ab} | 22.41 (4.33) ^a | 52.61 |
| Neem oil 2% | 39.78 (5.79) ^a | 12.33 (3.55) ^{cd} | 27.30 (5.12) ^{ab} | 37.96 (6.02) ^{ab} | 23.70 (4.78) ^a | 45.78 |
| NSKE 5% | 33.78 (5.83) ^a | 28.04 (5.11) ^{bc} | 23.15 (4.84) ^{ab} | 57.22 (7.26) ^a | 26.96 (5.05) ^a | 27.54 |
| Spiromesifen 240 SC @ 100g a.i. ha ⁻¹ | 21.96 (4.64) ^a | 2.67 (1.69) ^{de} | 3.33 (2.12) ^c | 13.26 (3.34) ^b | 10.66 (3.35) ^a | 83.98 |
| Fenazaquin 10 EC @ 125g a.i. ha ⁻¹ | 28.41 (5.37) ^a | 1.63 (1.38) ^c | 1.96 (1.60) ^c | 6.70 (2.63) ^b | 11.15 (3.39) ^a | 88.52 |
| Diafenthiuron 50 WP @ 400g a.i. ha ⁻¹ | 68.48 (7.83) ^a | 3.52 (1.98) ^{de} | 2.55 (1.36) ^c | 6.85 (2.83) ^b | 7.56 (2.71) ^a | 89.04 |
| Standard Check (Dicofol 18.5 EC @ 250g a.i. ha ⁻¹) | 32.34 (5.71) ^a | 2.55 (1.64) ^{de} | 2.67 (1.74) ^c | 6.56 (2.65) ^b | 10.52 (3.22) ^a | 88.06 |
| Untreated Control | 24.07 (4.85) ^a | 51.18 (7.18) ^a | 21.30 (4.76) ^{ab} | 51.18 (7.03) ^a | 63.15 (7.77) ^a | - |

DBT = Day Before Treatment; DAT = Days After Treatment ; Means followed by same letters do not differ significantly by DMRT ($p = 0.05$) ; Mean reduction- Mean reduction over untreated control; †- Values in the parenthesis are square root transformed values, ††- Values in the parenthesis are adjusted means of square root transformed values based on ANOCOVA

in case of NSKE 5 per cent (23.15) as against 27.30 in case of neem oil 2 per cent. There was no significant difference between the two acaropathogenic fungi, though *B. bassiana* recorded a lower population of 16.89 as compared to 36.56 mites per cm² leaf area recorded in case of *H. thompsonii*. The botanicals as well as the acaropathogenic fungi were on par with untreated control.

Similar trend was observed at ten days after spraying where the chemical acaricides continued to record lower mite populations with dicofol 18.5 recording the lowest count (6.56) as against 51.18 in untreated control. Fenazaquin 10 EC (6.70), diafenthiuron 50 WP (6.85) and spiromesifen 240 SC (13.26) were found to be on par with dicofol. All the other treatments viz., *H. thompsonii* (17.28), *B. bassiana* (30.63) neem oil 2 per cent (37.96) and NSKE 5 per cent (57.22) failed to record significant reduction in mite population as compared to untreated control.

At fourteen days after treatment, all the treatments were statistically on par with respect to mean mite population though diafenthiuron 50 WP recorded the lowest mite population of 7.56 per cm² leaf area.

Diafenthiuron 50 WP recorded the highest per cent reduction in the mite population over untreated control (89.04 %), closely followed by fenazaquin 10 EC (88.52 %) and dicofol 18.5 EC (88.06 %). The next best treatment was spiromesifen 240 SC with a mean per cent reduction of 83.98 over untreated control. Among the botanicals highest population reduction over control was exhibited by neem oil 2 per cent (45.78 %), followed by NSKE 5 per cent (27.54 %). *B. bassiana* reduced the mite population to the level of 52.61 per cent, followed by *H. thompsonii* (43.70 %).

4.4.1.2. Season II (November – February, 2013)

The results of the field experiment to evaluate the efficacy of different treatments against active stages of *T. urticae* are presented in Table 10. The mean

Table 10. Efficacy of treatments against active stages of *Tetranychus urticae* on okra during season II

| TREATMENTS | Mean no. of mites / cm ² leaf area | | | | | Mean reduction (%) |
|--|---|-------------------------------|-------------------------------|-------------------------------|------------------------------|--------------------|
| | 1 DBT† | 3 DAT†† | 7 DAT†† | 10 DAT†† | 14 DAT†† | |
| <i>Hirsutella thompsonii</i> @ 10 ⁷ spores/ml | 44.67 (6.70) ^a | 26.77 (4.69) ^{bc} | 29.30 (5.44) ^{ab} | 17.78 (4.21) ^{ab} | 9.15 (2.82) ^{bc} | 56.71 |
| <i>Beauveria bassiana</i> @ 10 ⁷ spores/ml | 71.48 (8.47) ^a | 31.00 (5.75) ^b | 26.41 (5.17) ^{ab} | 11.33 (3.30) ^{bc} | 7.81 (2.71) ^{bc} | 60.08 |
| Neem oil 2% | 64.71 (8.02) ^a | 18.89 (4.29) ^{cd} | 7.59 (2.83) ^c | 2.29 (1.65) ^d | 1.15 (1.25) ^d | 84.40 |
| NSKE 5% | 71.85 (8.29) ^a | 33.74 (5.89) ^b | 20.33 (4.53) ^b | 18.00 (4.29) ^{ab} | 12.22 (3.57) ^b | 56.04 |
| Spiromesifen 240 SC @ 100g a.i. ha ⁻¹ | 76.26 (8.75) ^a | 18.19 (4.49) ^{bc} | 1.67 (1.43) ^c | 2.41 (1.64) ^d | 2.82 (1.88) ^{cd} | 86.91 |
| Fenazaquin 10 EC @ 125g a.i. ha ⁻¹ | 63.86 (7.93) ^a | 17.63 (4.15) ^{cd} | 1.15 (1.24) ^c | 1.04 (1.24) ^d | 0.52 (0.99) ^d | 89.39 |
| Diafenthiuron 50 WP @ 400g a.i. ha ⁻¹ | 59.33 (7.59) ^a | 14.63 (3.60) ^{cd} | 1.78 (1.51) ^c | 1.15 (1.31) ^d | 3.03 (1.79) ^{cd} | 89.26 |
| Standard Check (Dicofol 18.5 EC @ 250g a.i. ha ⁻¹) | 69.07 (8.08) ^a | 8.11 (2.89) ^d | 5.81 (2.32) ^c | 3.37 (1.83) ^{cd} | 2.93 (1.73) ^{cd} | 89.45 |
| Untreated Control | 78.59 (8.80) ^a | 73.70 (8.84) ^a | 45.56 (6.58) ^a | 33.37 (5.67) ^a | 39.11 (6.38) ^a | - |

DBT = Day Before Treatment; DAT = Days After Treatment ; Means followed by same letters do not differ significantly by DMRT ($p = 0.05$) ; Mean reduction- Mean reduction over untreated control; †- Values in the parenthesis are square root transformed values, ††- Values in the parenthesis are adjusted means of square root transformed values based on ANOCOVA

population of active stages of *T. urticae* before imposing the treatments ranged from 44.67 to 78.59 per cm² leaf area.

At three days after treatment all the treatments significantly reduced the mite population as compared to untreated control. The lowest mite population of 8.11 per cm² leaf area was recorded in dicofol 18.5 EC followed by diafenthiuron 50 WP (14.63) and fenazaquin 10 EC (17.63). However these chemicals were also on par with each other. Spiromesifen 240 SC recorded an average of 18.19 mites per cm² leaf area and was on par with diafenthiuron 50 WP and fenazaquin 10 EC, but inferior to dicofol 18.5 EC. Among the botanicals, neem oil 2 per cent was found superior over NSKE 5 per cent and recorded a mite population of 18.89 as against 33.74 in the case of the latter. It was also on par with the chemicals fenazaquin 10 EC, spiromesifen 240 SC, diafenthiuron 50 WP and dicofol 18.5 EC. However both the botanicals were superior over untreated control which recorded a mean population of 73.70/cm² leaf area. The mean mite population was 26.77 in case of *H. thompsonii* as against 31.00 in *B. bassiana*. Both the acaropathogens and botanicals were on par with each other and superior over untreated control.

At seven days after spraying, all the treatments except acaropathogenic fungi caused significant reduction in mite population as compared to untreated control. The lowest mite population of 1.15 per cm² leaf area was recorded by Fenazaquin 10 EC which was also on par with spiromesifen 240 SC (1.67), diafenthiuron 50 WP (1.78) and dicofol 18.5 EC (5.81). Among the botanicals, neem oil 2 per cent recorded lower mite population of 7.59 compared to NSKE 5 per cent (20.33) and was also on par with the new acaricide molecules and standard check. However both the botanicals were superior over untreated control (45.56). The acaropathogens, *H. thompsonii* (29.30) and *B. bassiana* (26.41) were found to be on par with each other and untreated control.

Similar trend was observed at ten days after spraying where the chemical molecules continued to record lower mite populations with fenazaquin 10 EC recording the lowest mite population of 1.04 per cm² leaf area. It was also found on par with diafenthiuron 50 WP (1.15), spiromesifen 240 SC (2.41) and standard check dicofol 18.5 EC (11.93). A mean mite population of 2.29 was recorded by Neem oil 2 per cent which was superior over NSKE 5 per cent (18.00). *B. bassiana* which recorded a mean mite population of 11.33 as against 17.78 in *H. thompsonii* was on par with the same. However *H. thompsonii* was also on par with untreated control (33.37).

At fourteen days after spraying, all the treatments recorded significant reduction in mite population as compared to untreated control. The new molecules, fenazaquin 10 EC, diafenthiuron 50 WP, spiromesifen 240 SC and dicofol 18.5 EC continued to record lower mite population per cm² leaf area. The lowest mean mite population was recorded by fenazaquin 10 EC (0.52) which was on par with spiromesifen 240 SC (2.82), diafenthiuron 50 WP (3.03) and dicofol 18.5 EC (2.93). Among the botanicals, neem oil 2 per cent recorded lower mite population (12.22) as compared to NSKE 5 per cent (12.22) and was superior over the same. Both the acaropathogenic fungi were found superior to untreated control and recorded a mean mite population of 7.81 in *B. bassiana* and 9.15 per cm² leaf area in *H. thompsonii*.

The highest reduction in mite population over untreated control was observed in dicofol 18.5 EC (89.45 %), fenazaquin 10 EC (89.39 %) and diafenthiuron 50 WP (89.26 %). The next best treatments with respect to per cent reduction in mite population were spiromesifen 240 SC (86.91 %) and neem oil 2 per cent (84.40 %) as against 56.04 per cent in NSKE 5 per cent. The acaropathogens recorded mite population to the tune of 60.08 per cent in case of *B. bassiana* and 56.71 per cent in case of *H. thompsonii*.

Discussion

5. DISCUSSION

The present study was undertaken to investigate the biology of two-spotted spider mite, *Tetranychus urticae* Koch and to evaluate the efficacy of acaropathogenic fungi, botanicals and new acaricide molecules against *T. urticae* on okra. The observations and inferences based on the study are discussed below in the light of available literature.

5. 1. Biology of *Tetranychus urticae* Koch on okra

The studies on the biology of *T. urticae* were conducted in the Acarology laboratory at $30 \pm 3^{\circ}\text{C}$ and $61.5 \pm 7\%$ relative humidity on leaf discs of okra. The developmental stages of *T. urticae* on okra consisted of egg, larva, protonymph, deutonymph and adult stage as reported by Rajkumar (2003) in jasmine and Silva *et al.* (2009) in gerbera. Mallick and Channabasavanna (1983) as well as Patil (2005) have reported similar finding in *T. ludeni* infesting French bean plant and *T. macfarlanei* on brinjal, indicating the uniformity of developmental pattern for the family Tetranychidae.

The development of egg, larva and nymphs of *T. urticae* on okra in the present study was shorter (Fig. 1) compared to that on jasmine as reported by Rajkumar (2003). In jasmine the incubation period of *T. urticae* was more than four days in both males and females and the mite took a minimum of one day for completing each of the developmental stages from larva to adult emergence. Similar observations were also made by Silva *et al.* (2009) on *T. urticae* infesting gerbera. The influence of host plant on the developmental duration was discussed by several workers in different species of *Tetranychus*. When the biology of *T. urticae* was studied on six common bean cultivars in Iran, significant variation was observed in terms of total developmental period of males and females (Najafabadi, 2012). Adango *et al.* (2006) on studying the biology of *T. ludeni* on two leafy vegetables,

found that the developmental duration of *T. ludeni* was shorter on amaranth than on nightshade, confirming the role of host plant on the biology of *T. urticae*. Naher *et al.* (2008) observed that the life cycle of *T. urticae* was completed within 4.22 days at 28.53°C but only in 28.33 days at 13.78 °C, which is again in agreement with the findings of the present study where the males and females attained their adulthood in a short period of 6.73 and 7.52 days at 30°C. The higher temperature prevalent during the study period could have accelerated the developmental rate and reduced the duration of development. The quiescent intervals such as nymphochrysalis, deutochrysalis and teleiochrysalis recorded in the study have been reported by Riahi *et al.* (2011) and Naher *et al.* (2008) in *T. urticae* infesting peach and country bean and in *T. macfarlanei* infesting brinjal (Patil, 2005).

Difference in duration of development could also be influenced by sex. For instance, males of *T. urticae* recorded a shorter developmental period of 6.73 days compared to 7.52 days in females on okra (Fig. 2). Several workers had already reported a similar trend in the developmental biology of different species of *Tetranychus* spp. infesting various crops. El-Wahed and El-Halawany (2012) reported that males of *T. urticae* completed its life cycle in 6.3 days and females in 6.5 days on pear, while Rajkumar (2003) observed a duration of 10.70 days for male and 12.36 days for female to complete their life cycle in jasmine. Manjunatha and Puttaswamy (1989) observed that females and males of *T. neocaledonicus* took 10.44 days and 10.19 days to complete their life cycle on French bean plants. Sathiamma (1991) also reported similar results for *T. ludeni*. The early emergence of male ensures sexual reproduction and sustenance of population as against parthenogenesis which is commonly observed in *T. urticae* in the absence of males and which results in all male population.

The deutonymphs and adults of female *T. urticae* were larger in size compared to males as also reported by Patil (2005) in case of *T. macfarlanei*. Adult

Figure 1. Duration of development stages of *T. urticae*

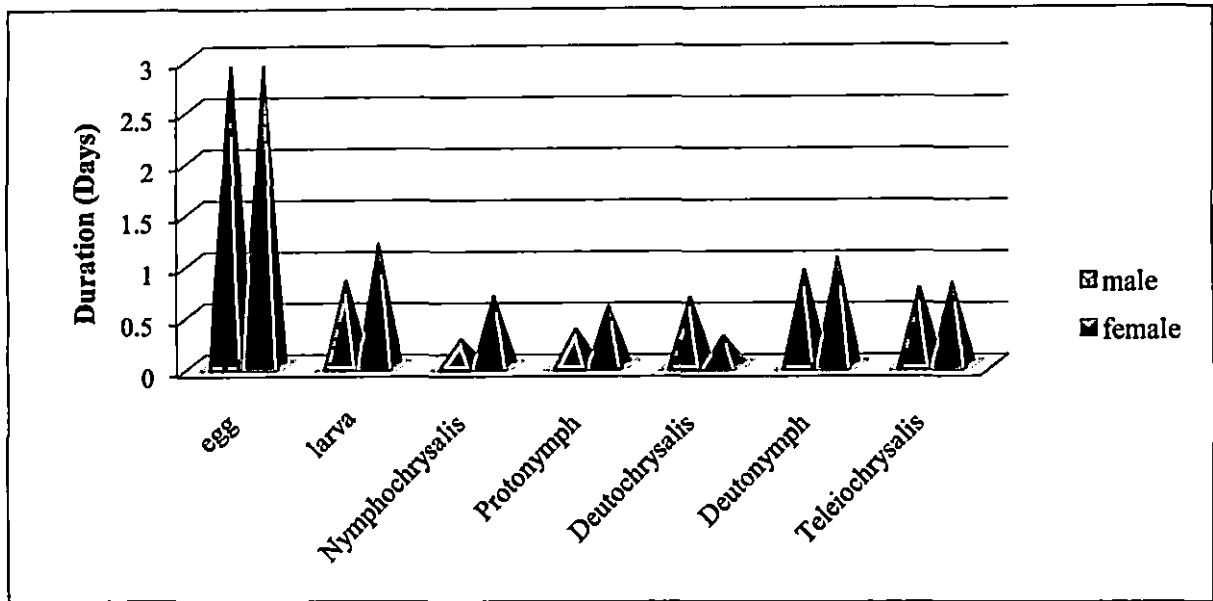


Figure 2. Total development period of *T. urticae*

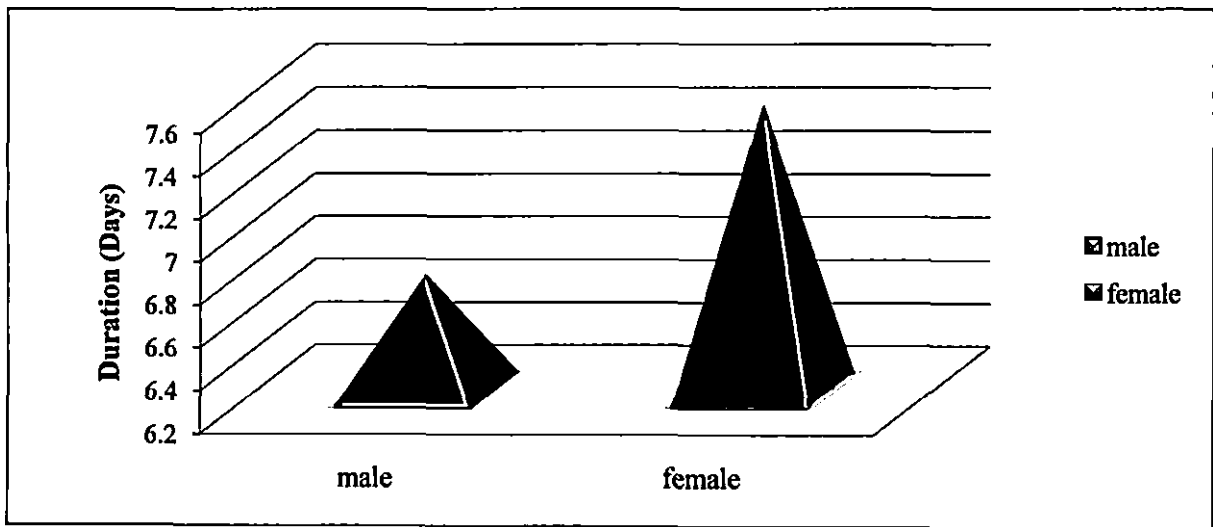


Figure 1. Duration of development stages of *T. urticae*

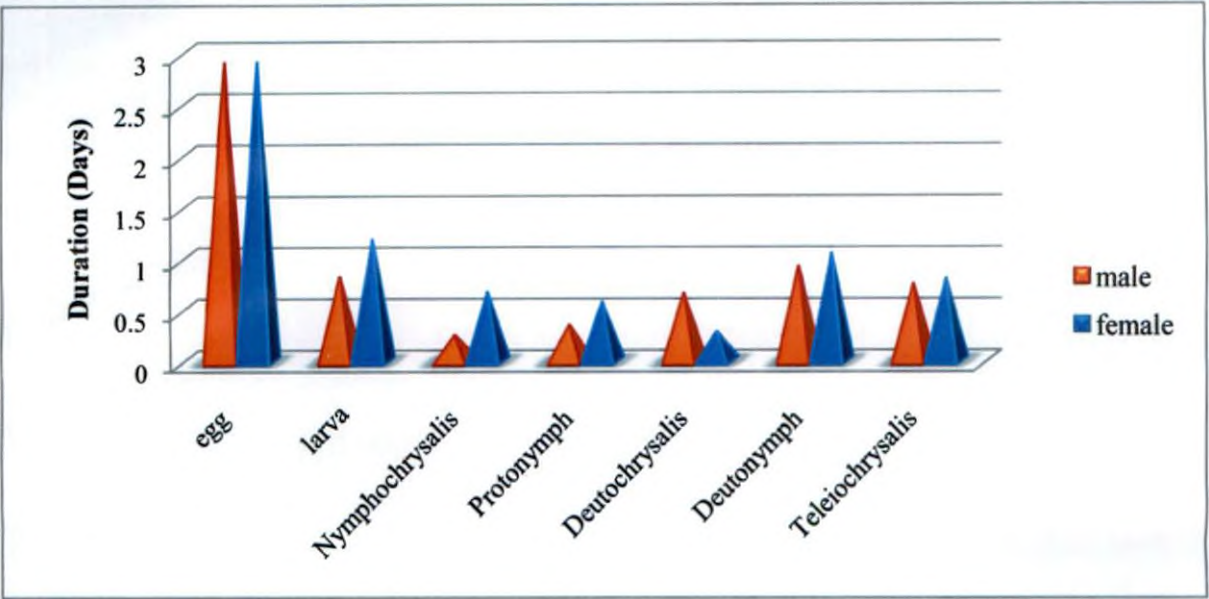
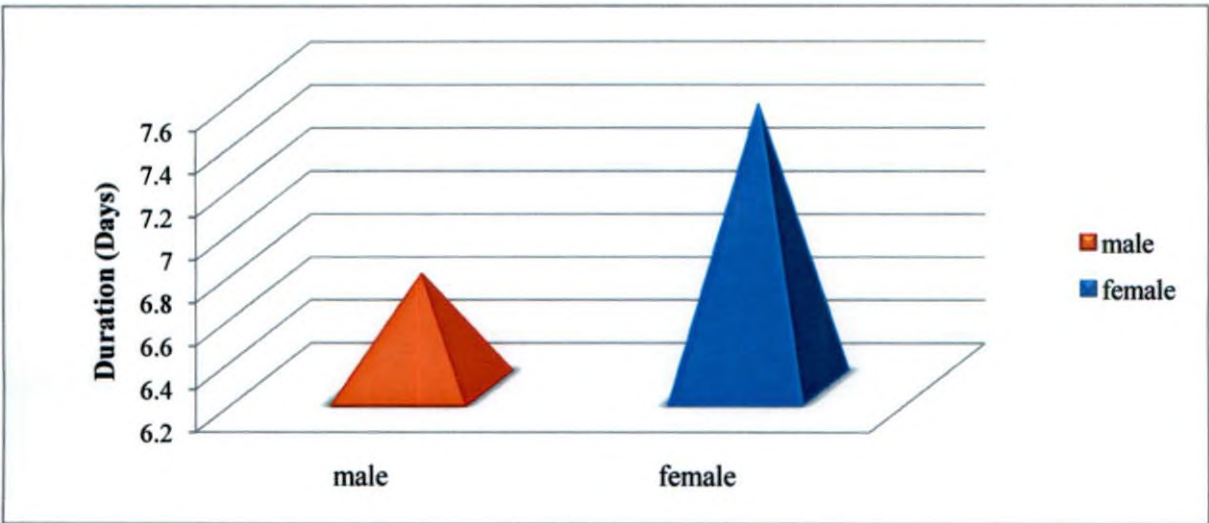


Figure 2. Total development period of *T. urticae*



male was reddish green or light red in colour with the body tapering posteriorly to a blunt point, whereas the females were larger, plumper and carmine red in colour with longer setae all over the body. Similar sexual dimorphism was also reported by Seeman and Beard (2011) in *T. urticae*, Patil (2005) in *T. macfarlanei* and Kaimal and Ramani (2011) in *T. ludeni*.

The males rested over the quiescent female deutonymphs which they came across to mate immediately after the emergence of the female. This peculiar behaviour prior to mating was common among *T. urticae* (Seeman and Beard, 2011) and other spider mites (Kaimal and Ramani, 2011; Manjunatha and Puttaswamy, 1989). Male pushed and raised the posterior abdominal region of the female and slide underneath with its hysterosoma upturned (Rajkumar, 2003; Patil, 2005).

T. urticae preferred to colonize and lay eggs on the underside of leaves. Gravid female of *T. urticae* laid eggs randomly on both the webbings as well as on the leaves. This behaviour was also observed in *T. ludeni* (Puttaswamy and Channabasavanna, 1980; Mallik and Channabasavanna, 1983). *T. urticae* showed a general preference to lay eggs near the veins and midrib of okra. The preference for the under surface of the leaves for oviposition could easily be due to the better protection from direct sunlight and rainfall. However, Kaimal and Ramani (2011) noticed that *T. ludeni* showed no such preference between the leaf surfaces for oviposition.

Mated females produced a progeny consisting of both males and females in the ratio 1: 5.8. Kaimal and Ramani (2011) reported a similar sex ratio of 1:5 in *T. ludeni* on velvet bean whereas Manjunatha and Puttaswamy (1989) reported a wider sex ratio (1:10) in *T. neocaledonicus* on French beans. As the males are known to be polygamous (Kaimal and Ramani, 2011; Patil, 2005), the chances of females getting fertilized become high which will ensure population build up and sustenance of the species.

Unmated females of *T. urticae* exhibited arrhenotokous reproduction. This is in conformity with earlier reports by Sabelis (1981) in *T. urticae* and in other species of *Tetranychus* by Manjunatha and Puttaswamy (1989) as well as Bonato and Gutierrez (1999). This behaviour of unmated females to produce a progeny of males may be for imparting a natural control over its population when the food source is scarce or space is limited, thereby increasing the chances for survival.

Mated females laid more number of eggs (108 eggs) compared to unmated females (77 eggs). This higher fecundity of mated females is in accordance with the findings of Rajkumar (2003) in *T. urticae*. However Kaimal and Ramani (2011) and Kaimal *et al.* (2007) reported that in *T. ludeni* the unmated females recorded higher number of eggs. Effect of temperature on fecundity of the mite has been extensively studied by various workers. Rate of reproduction was significantly increased as the temperature increased with highest mean fecundity of 156.8 eggs at 30°C (El-Wahed and El-Halawany, 2012). This might be the reason for recording a high fecundity of 108 eggs at 30°C in the present study. The higher egg viability of 92.10 per cent observed in the current study, is an evidence of the high biotic potential of this species. Silva *et al.* (2009) had reported a higher egg viability of 96.5 per cent in *T. urticae* on gerbera.

In the present study, the longevity of unmated females was more than that of mated females and males as already reported by Puttaswamy and Channabasavanna (1980) in *T. ludeni*. The negative influence of mating on life expectancy was also reported by Puttaswamy and Reddy (1980) and Manjunatha and Puttaswamy (1989) in other species on tetranychid mites as well.

The shorter developmental period of *T. urticae* averaging 6-8 days and high fecundity of 108 eggs observed in the present study would help them in successfully completing 2-5 generations per month at $30 \pm 3^{\circ}\text{C}$ and $61.5 \pm 7\%$ relative humidity. The shortening of life cycle along with significant increase in the rate of

reproduction in *T. urticae* with the increase in temperature was reported by several workers (Fahnbulleh, 2007; El-Wahed and El-Halawany, 2012; Riahi *et al.*, 2013). This explains why this mite could multiply and attain pest status especially during the drier and hotter months of the year in Kerala (Kaimal, 2009).

5.2. Evaluation of the efficacy of selected acaropathogenic fungi, botanicals and new acaricide molecules against the two-spotted spider mite *T. urticae* on okra

Field study was conducted to evaluate the efficacy of two acaropathogenic fungi, two botanicals and three new acaricide molecules along with a standard check and an untreated control against the two-spotted spider mite on okra in the experimental plot at College of Horticulture, Vellanikkara during two seasons *viz.*, February – May, 2012 and November, 2012 – February, 2013.

Though the new acaricide molecules, diafenthiuron 50 WP, fenazaquin 10 EC, spiromesifen 240 SC showed a similar trend during the two seasons of study, the acaropathogenic fungi and botanicals differed in their performances in the two seasons tested against *T. urticae* and hence could not obtain consistent results. This may be due to the environmental factors that modify the microclimate of the crop, either favouring or adversely affecting the performances of the same. So the treatment effects are discussed separately during the two seasons of study.

Spiromesifen, a tetrone acid derivative acts as inhibitor of acetyl-CoA-carboxylase, a key enzyme in fatty acid biosynthesis. It is highly toxic to eggs and immature stages of spider mites, while it acts more slowly against adult females, causing reduction in fertility and fecundity (Marcic *et al.*, 2011). In baseline susceptibility studies conducted by Nauen *et al.* (2005), spiromesifen did not have a marked effect against *T. urticae* adult females but was highly toxic against eggs of the mite. Sato *et al.* (2011) observed that among the different developmental stages studied, the egg stage of *T. urticae* was found to be the most sensitive to

spiromesifen. In the present study also spiromesifen recorded a higher reduction in egg count over untreated control (Fig. 8) compared to standard check, dicofol. Spiromesifen has been reported to show better efficacy than dicofol against phytophagous mites by Kavitha *et al.* (2006). During the two seasons of study, spiromesifen consistently recorded lower egg and mite counts up to fourteen days (Fig. 3, 4, 6 and 7). Sato *et al.* (2011) reported that under field condition, a complete suppression of population of *T. urticae* could be achieved in ten days time using spiromesifen. In capsicum spiromesifen proved to be very effective against mites, bringing remarkable control for approximately one month after application (Fanigliulo *et al.*, 2010) revealing its good ovicidal action and residual effect. Saryazdi *et al.* (2013) observed ovicidal activity as well as reduction in the survival rate, fecundity and egg hatching rate when spiromesifen was used. This peculiar growth regulatory effect of spiromesifen might have contributed to its high efficacy in the field as observed in the present study.

It is evident from Tables 7, 8, 9 and 10 that diafenthiuron 50 WP, fenazaquin 10 EC, spiromesifen 240 SC and dicofol 18.5 EC were significantly superior over acaropathogenic fungi and botanicals, bringing down the population up to 14 days of treatment application. The effectiveness of newer molecules against tetranychid mites have been extensively studied by several workers.

The insecto-acaricide diafenthiuron, is a novel thiourea compound that disrupts oxidative phosphorylation by inhibition of the mitochondrial ATP synthase enzyme. It has been reported as effective against active stages of spider mites (Marcic *et al.*, 2011). Similar results were also reported by Patil, (2005) who found that use of diafenthiuron resulted in more than 96 per cent mortality of adult mites. The ovicidal activity of diafenthiuron was evaluated by Patil and Nandihalli (2007) who, based on their bioassay studies on *T. macfarlanei* infesting brinjal, reported that diafenthiuron caused more than 98 per cent egg mortality. Onkarappa and

puttaswamy, 1999) reported that under polyhouse condition diafenthiuron (300, 450 and 600g a.i. ha⁻¹) could effectively reduce the population of eggs, nymphs and adults of *T. urticae* infesting okra which supports the findings of the present study. Brits and Vickers (1990) observed that diafenthiuron could control *T. cinnabarinus* in cotton for up to 8 days. Diafenthiuron 50 WP offered maximum control over spider mites up to 7 days of treatment application (AINPAA, 2011). The findings of the present study, where mite population was kept under check for up to seven and ten days in season 1 and season 2 respectively are in agreement with the above studies (Fig. 4 and 7). In both seasons, diafenthiuron consistently recorded higher per cent reduction in mite and egg counts over untreated control (Fig.5 and 8) showing its high efficacy against all stages of the mite. The residual activity of diafenthiuron combined with its effect on eggs and motile stages of mite may be the reason for obtaining long term control over the mite population in the present study.

Fenazaquin is an acaricide which belongs to quinazoline class of chemicals which inhibits mitochondrial electron transport (MET) at complex I. It has high efficacy against eggs and motile stages of tetranychid mites (Marcic *et al.*, 2011). In a laboratory bioassay conducted to test the ovicidal activity against *T. urticae* on okra, Sangeetha and Ramaraju (2013) reported 81.25 per cent egg mortality for fenazaquin 10 EC at 125g a.i. ha⁻¹. Fenazaquin 10 EC was also reported to cause 90.52 per cent mortality of adult mites of *T. macfarlanei* by Patil in 2005. In the present study, fenazaquin recorded significantly lower mite population for up to fourteen days of treatment (Fig. 4 and 7). It also recorded a considerable reduction in egg and mite population over untreated control (Fig. 5 and 8). In strawberry, fenazaquin was found to cause significantly higher mortality of *T. urticae* three days after the spray. After seven days, the same proved highly effective by maintaining the mite population under control, which extended up to 15 days of application (AINPAA, 2011). A similar trend in the efficacy of Fenazaquin was evidenced in the present study also.

Figure 3. Effect of different treatments on egg stage of *T. urticae* during season I

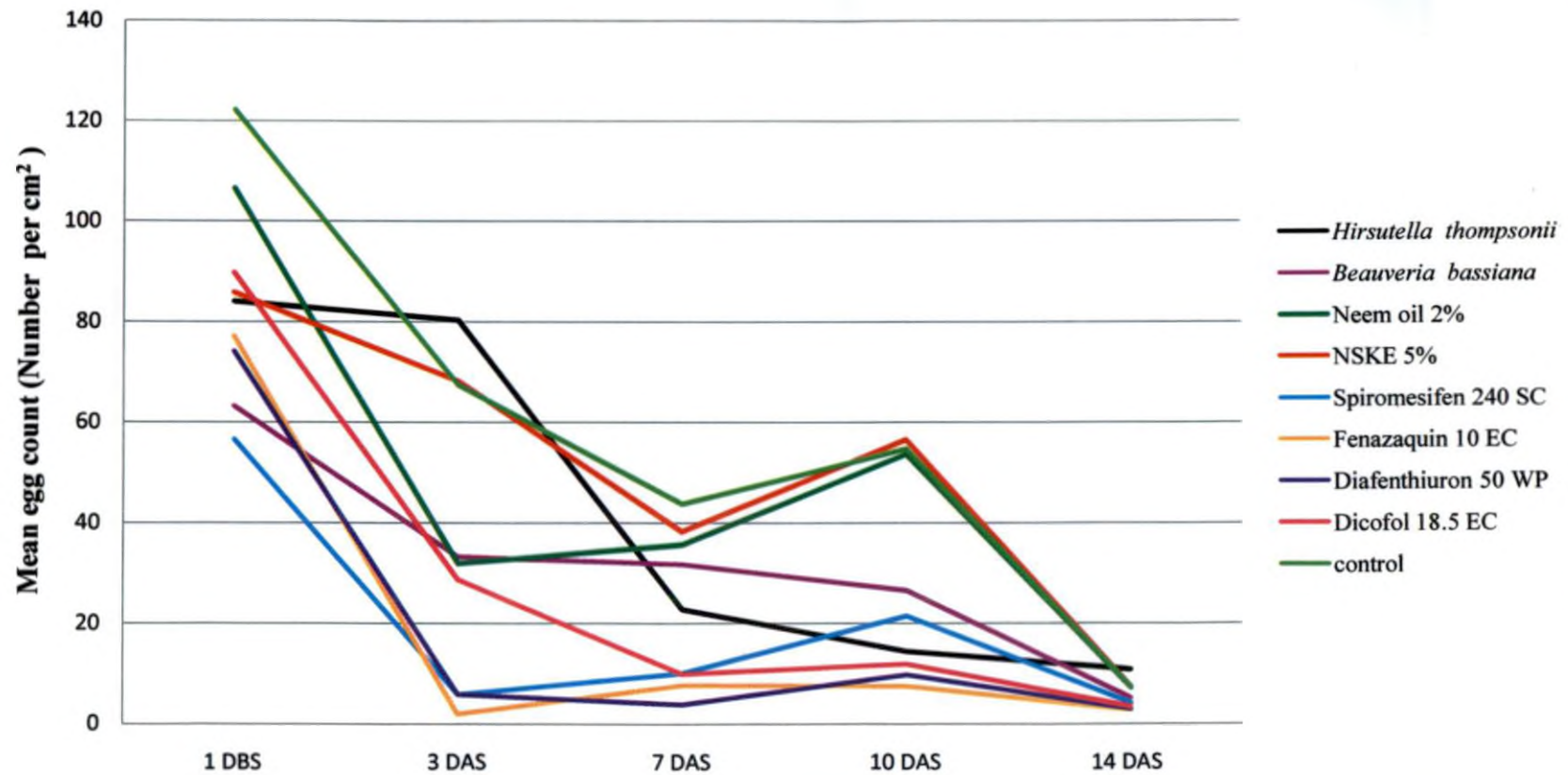


Figure 4. Effect of different treatments on the population of active stages of *T. urticae* during season I

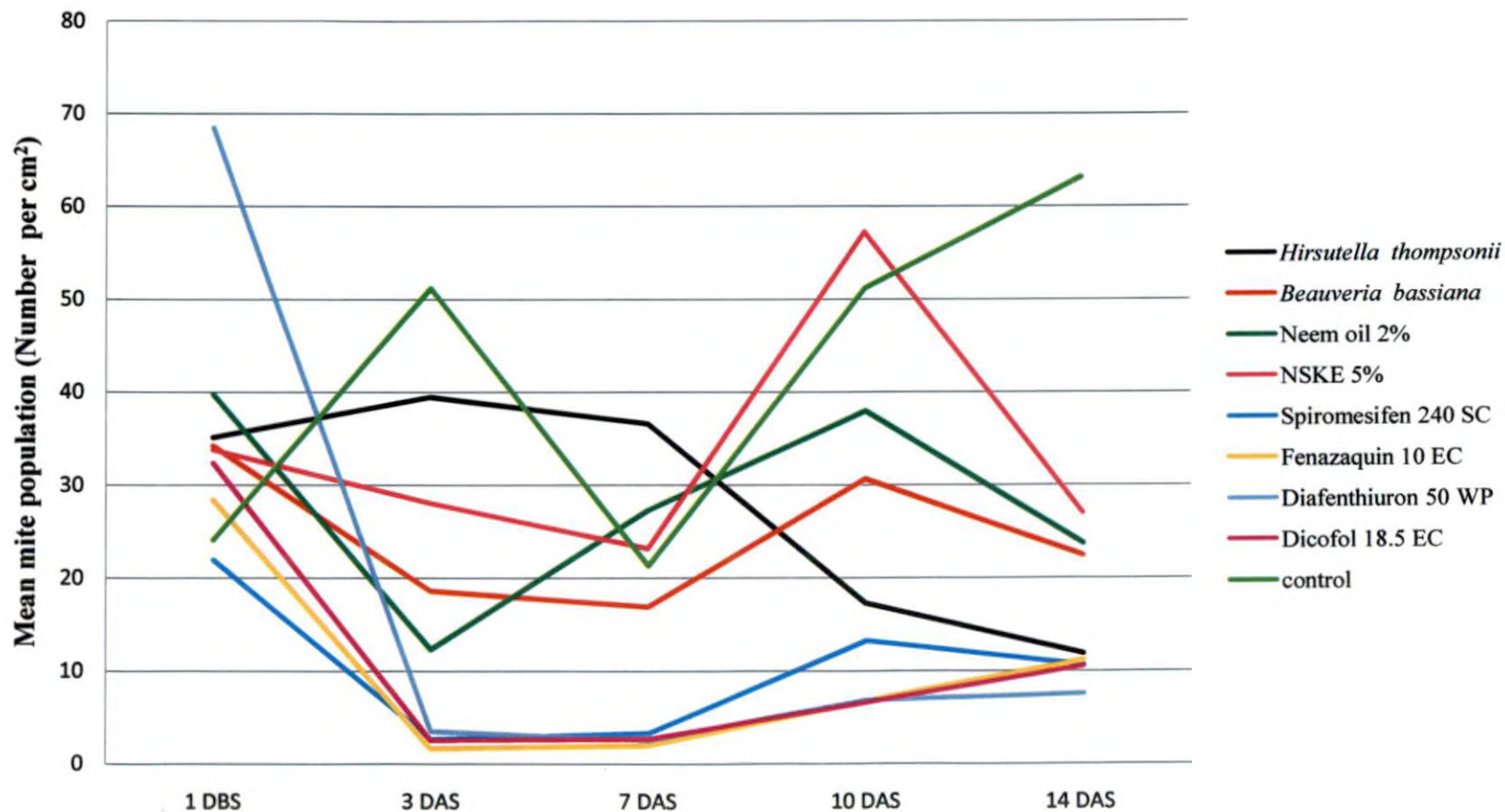


Figure 5. Field efficacy of different treatments against *T. urticae* on okra during season I

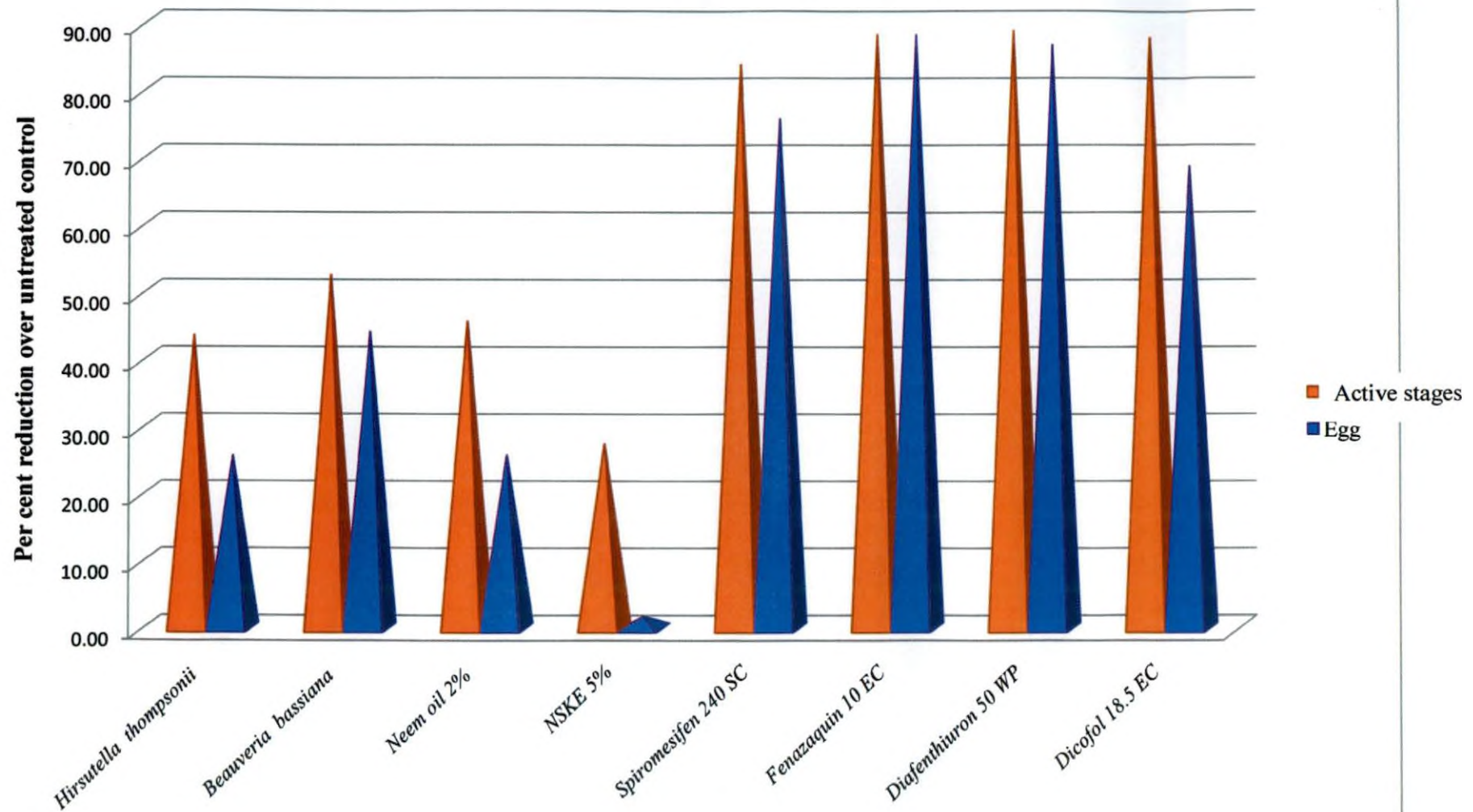


Figure 6. Effect of different treatments on egg stage of *T. urticae* during season II

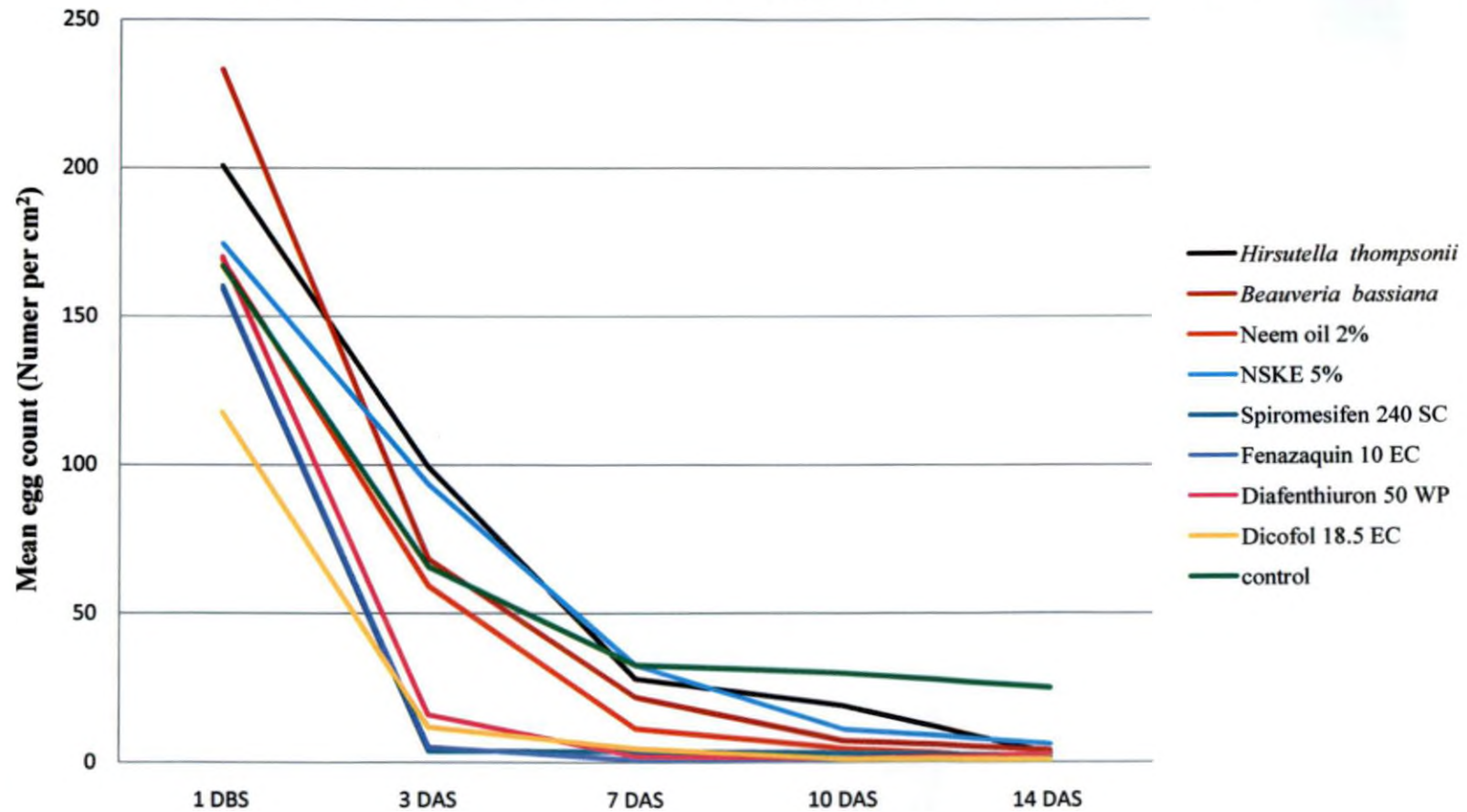
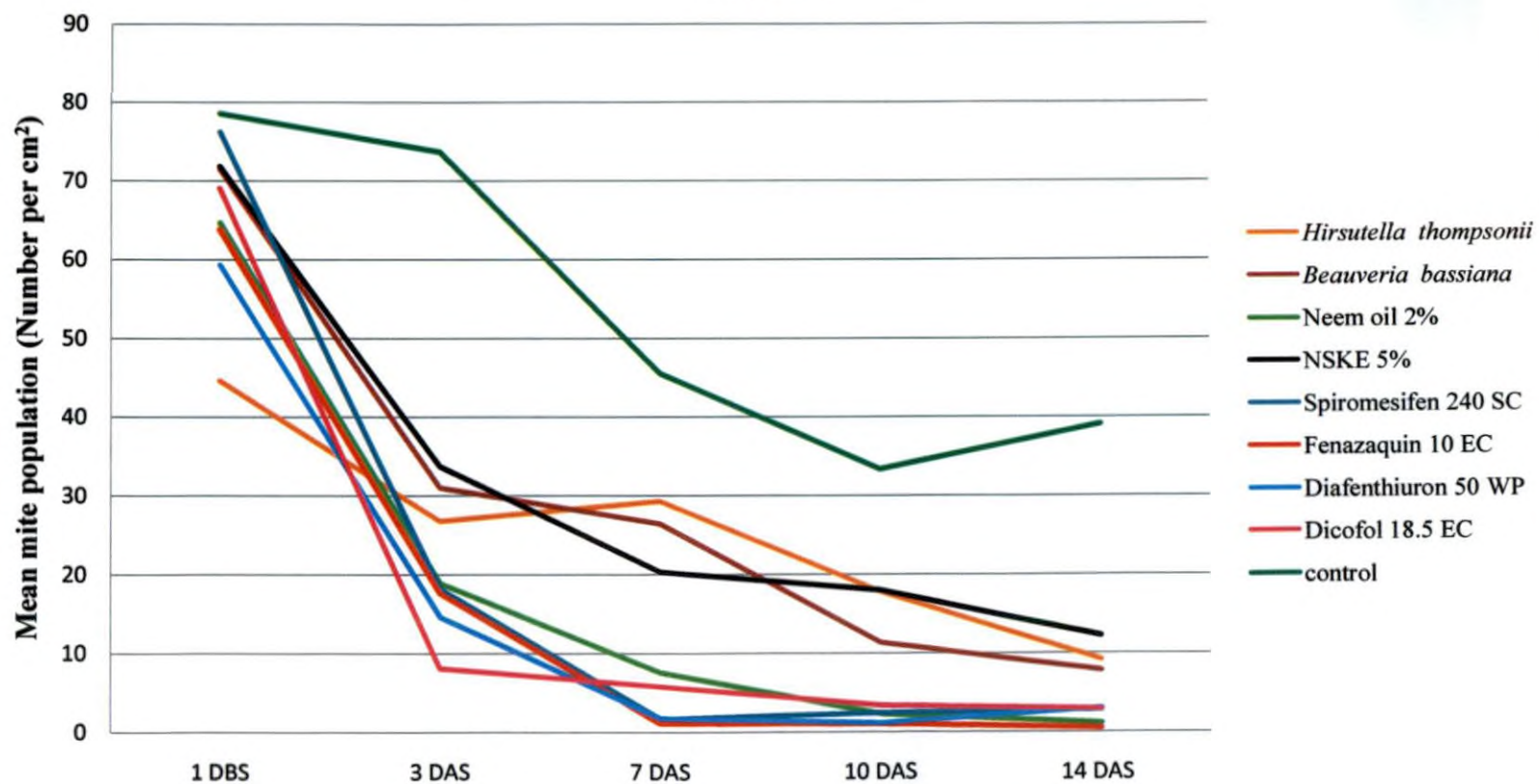
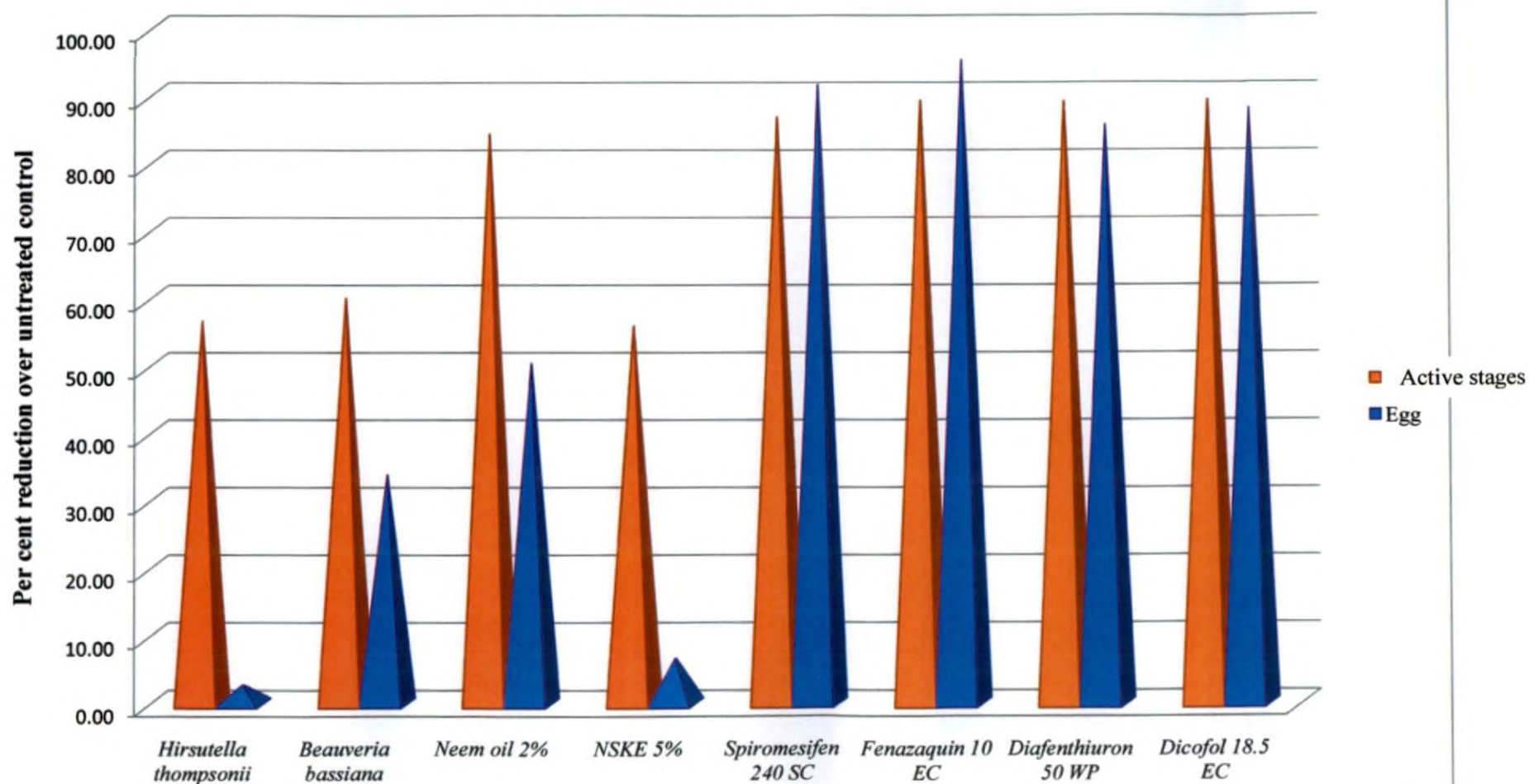


Figure 7. Effect of different treatments on the population of active stages of *T. urticae* during season II



Active stages

Figure 8. Field efficacy of different treatments against *T. urticae* on okra during season II



In the present study, new acaricide molecules have exhibited acaricidal activity on par with dicofol 18.5 EC (Fig 5 and 8). This indicates that in the present scenario of facing out of toxic chemicals which are not safe to non target organisms and environment, these new molecules can substitute dicofol with rotational use to avoid development of resistance and ill effects on environment.

Among the botanicals evaluated, the acaricidal activity of neem oil 2 per cent and NSKE 5 per cent on motile stages of *T. urticae* lasted only up to three days of treatment during season1 (Fig. 4). Further, there was no significant reduction in egg count over untreated control. However, both neem oil 2 per cent and NSKE 5 per cent were found to perform better during season 2. The acaricidal activity of neem oil was superior to that of NSKE and was on par with that of chemicals (Fig. 5 and 8). The acaricidal activity of neem oil 2 per cent and NSKE 5 per cent on motile stages of *T. urticae* lasted up to fourteen days of treatment application though the effect of NSKE 5 per cent was found to decrease from tenth day onwards. A significant reduction in egg count was caused by neem oil 2 per cent from seven to fourteen days after spraying where as NSKE 5 per cent showed its superiority over control from tenth day onwards (Fig. 6).

This varied effect of neem based botanicals on eggs and active stages of mite may be attributed to slow action of azadirachtin, which includes complete or partial antifeedant response, delayed and/or disrupted moulting and inhibited reproduction (Copping and Duke, 2007). The studies on spider mites indicate that azadirachtin, in addition to being toxic to various development stages, acts as antifeedant, reduces fecundity and fertility and shortens the life span of adult mite (Sundaram & Sloane, 1995). Patil and Nandihalli (2009) reported that neem oil 2 per cent exhibited maximum acaricidal action against red spider mite in brinjal. Among many botanicals tested in the polyhouse against *T. urticae* on rose, neem oil 2 per cent was found to be the most effective while NSKE 5 per cent was only moderately effective

(Kumar, 2007) as also found in the second season trial of present study. Since no consistent results could be drawn on these botanicals from the present study, further trials should be undertaken to obtain conclusive results.

Hirsutella thompsonii is an acarine mycopathogen belonging to the class deuteromycetes. It penetrates the mite's integument mainly through the legs and forms hyphal bodies in chains in the haemolymph. Hyphae on which the spores were produced emerged through the genital and anal apertures and then all over the body (Gerson *et al.*, 1979). *Beauveria bassiana*, is a filamentous entomopathogenic fungus, belonging to the class deuteromycetes. It causes white muscardine disease by invading directly through the cuticle (Sandhu *et al.*, 2012).

During the present study, a significant reduction in the egg count and mite population was observed ten to fourteen days after application of the fungal pathogens *H. thompsonii* and *B. bassiana*, which shows that the bioagents require more time for their development on the host and finally cause mycosis under field conditions (Fig. 3, 4, 6 and 7). Kalmath *et al.* (2008), who evaluated the efficacy of various fungal isolates in the green house, observed that nine days after the first spray treatments *B. bassiana* alone and *B. bassiana* in combination with *H. thompsonii* recorded significant reduction in mite population over untreated control. In the glasshouse trial conducted on French bean against red spider mite, Ghosh *et al.* (2007) reported that *H. thompsonii* recorded lower mite population ten days after treatment application which supports the present study. During both the seasons of study comparatively lower performance was observed in case of these pathogens, (Fig. 5 and 8) indicating the influence of microclimatic conditions on the survival, development and initiation of epizootics by these fungi. The prevalence of high temperature during the period of study (Appendix I) might have had a negative influence on the same which has been observed in previous studies as well (Bugeme

et al., 2008; Gerson, *et al.*, 1979). As a result, no consistent results have been obtained on their efficacy against *T. urticae* in the field.

Summary

6. SUMMARY

The study entitled “Biology and management of the two-spotted spider mite, *Tetranychus urticae* Koch (Prostigmata: Tetranychidae) on okra, [*Abelmoschus esculentus* (L.) Moench]” was carried out at the Department of Agricultural Entomology, College of Horticulture, Vellanikkara during February, 2012 – June, 2013 to investigate the biology of two-spotted spider mite, *Tetranychus urticae* Koch and evaluate selected acaropathogenic fungi viz., *Hirsutella thompsonii* and *Beauveria bassiana*, botanicals viz., neem oil 2 per cent and NSKE 5 per cent, and new acaricide molecules viz., spiromesifen 240 SC, fenazaquin 10 EC and diafenthiuron 50 WP against *T. urticae* on okra.

The salient findings of the study are summarized hereunder.

- ▲ The biology of *T. urticae* was studied under laboratory conditions during October-November, 2012 at $30 \pm 3^{\circ}\text{C}$ and $61.5 \pm 7\%$ relative humidity. The life cycle of *T. urticae* composed of egg, larva, protonymph, deutonymph and adult. The immature stages viz., larval and nymphal stages were followed by short quiescent intervals called nymphochrysalis, deutochrysalis and teleiochrysalis respectively.
- ▲ The mite recorded an incubation period of 2.92 days. Larval period of 0.83 and 1.19 days, protonymphal period of 0.36 and 0.58 days and deutonymphal period of 0.67 and 0.29 days were recorded respectively, in male and female *T. urticae*.
- ▲ The total developmental period from egg to adult emergence was shorter for male (6.73 days) compared to female (7.52 days). Male was reddish green or light red in colour, smaller in size with body tapering posteriorly to a blunt point. Female was carmine red in colour, larger and plumper with longer setae over the body and legs.

- ^ The adult mite recorded longevity of 12, 12.5 and 17 days for male, mated female and unmated female, respectively.
- ^ *T. urticae* exhibited both sexual and parthenogenetic reproduction. Mated female's progeny consisted of both males and females in the ratio 1:5.8, whereas unmated female produced 100 per cent males.
- ^ Pre-oviposition, oviposition and post-oviposition periods lasted for 0.58, 9 and 4 days and 0.58, 11 and 4.5 days, respectively in mated and unmated female. Mated and unmated females on an average produced 108 and 77 eggs.
- ^ Field study was conducted to evaluate two acaropathogenic fungi (*Hirsutella thompsonii* and *Beauveria bassiana*), two botanicals (Neem oil 2 per cent and NSKE 5 per cent) and three new acaricide molecules (Spiromesifen 240 SC, Fenazaquin 10 EC and Diafenthiuron 50 WP) along with a standard check (Dicofol 18.5 EC) and an untreated control against *T. urticae* on okra during two seasons viz., February – May, 2012 and November, 2012 – February, 2013. The new acaricide molecules diafenthiuron 50 WP, fenazaquin 10 EC and spiromesifen 240 SC exhibited high efficacy against various stages of *T. urticae* and significant reduction in egg and mite population was observed fourteen days after spraying.
- ^ Among the botanicals evaluated, neem oil 2 per cent was found to be superior to NSKE 5 per cent during the second season though on par with each other during the first season of study. Among the acaropathogenic fungi, *B. bassiana* was found to perform better than *H. thompsonii* in terms of per cent reduction over untreated control. However consistent results are lacking for bioagents and botanicals.

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Appendix

**Appendix I. Weekly meteorological data recorded at College of Horticulture,
Vellanikkara during 2012- 2013**

| Month/week | Temperature (°C) | | Relative humidity (%) | Rainfall (mm) |
|-----------------------------------|------------------|---------|-----------------------|---------------|
| | Maximum | Minimum | | |
| 2012 | | | | |
| February I (5/2/12-11/2/12) | 34.7 | 23.0 | 59 | 0 |
| February II (12/2/12-18/2/12) | 35.1 | 22.5 | 61 | 0 |
| February III (19/2/12-25/2/12) | 35.9 | 21.0 | 48 | 0 |
| February IV (26/2/12-4/3/12) | 35.6 | 22.2 | 59 | 0 |
| March I (5/3/12-11/3/12) | 33.8 | 23.2 | 69 | 0 |
| March II (12/3/12-18/3/12) | 36.0 | 25.0 | 67 | 1 |
| March III (19/3/12-25/3/12) | 35.2 | 24.4 | 64 | 0 |
| March IV (26/3/12-1/4/12) | 35.8 | 24.9 | 69 | 0 |
| April I (2/4/12-8/4/12) | 35.1 | 24.2 | 71 | 1 |
| April II (9/4/12-15/4/12) | 35.8 | 24.8 | 69 | 0 |
| April III (16/4/12-22/4/12) | 35.4 | 25.1 | 71 | 2 |
| April IV (23/4/12-29/4/12) | 33.2 | 24.7 | 79 | 4 |

| | | | | |
|-------------------------------------|------|------|----|---|
| April V (30/4/12-6/5/12) | 32.4 | 25.3 | 77 | 1 |
| May I (7/5/12-13/5/12) | 32.9 | 25.1 | 76 | 1 |
| May II (14/5/12-20/5/12) | 33.2 | 25.2 | 77 | 2 |
| May III (21/5/12-27/5/12) | 32.2 | 25.4 | 79 | 1 |
| May IV (28/5/12-27/5/12) | 32.5 | 25.9 | 75 | 1 |
| November I (5/11/12-11/11/12) | 32.8 | 23.4 | 75 | 1 |
| November II (12/11/12-18/11/12) | 32.7 | 21.9 | 63 | 0 |
| November III (19/11/12-25/11/12) | 32.7 | 23.0 | 75 | 1 |
| November IV (26/11/12-2/12/12) | 33.1 | 22.0 | 53 | 0 |
| December I (3/12/12-9/12/12) | 33.2 | 23.6 | 63 | 1 |
| December II (10/12/12-16/12/12) | 33.3 | 21.5 | 58 | 0 |
| December III (17/12/12-23/12/12) | 31.6 | 24.2 | 55 | 0 |
| December IV (24/12/12-31/12/12) | 33.2 | 23.6 | 59 | 1 |
| | | | | |

| 2013 | | | | |
|----------------------------------|------|------|----|---|
| January I (1/1/13-7/1/13) | 34.4 | 23.0 | 61 | 0 |
| January II (8/1/13-14/1/13) | 33.9 | 23.0 | 52 | 0 |
| January III (15/1/13-21/1/13) | 33.5 | 21.0 | 48 | 0 |
| January IV (22/1/13-28/1/13) | 34.5 | 22.2 | 41 | 0 |
| January V (29/1/13-4/2/13) | 34.2 | 23.5 | 51 | 0 |
| February I (5/2/13-11/2/13) | 35 | 23.6 | 61 | 0 |
| February II 12/2/13-18/2/13 | 34.6 | 24.4 | 61 | 1 |
| February III 19/2/13-25/2/13 | 33.9 | 23.0 | 58 | 1 |
| February IV 26/2/13-4/3/13 | 36.3 | 21.8 | 42 | 0 |

**BIOLOGY AND MANAGEMENT OF THE TWO-
SPOTTED SPIDER MITE, *TETRANYCHUS URTICAE*
KOCH (PROSTIGMATA: TETRANYCHIDAE) ON OKRA
[*ABELMOSCHUS ESCULENTUS* (L.) MOENCH]**

By

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(2011-11-117)

ABSTRACT OF THE THESIS

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ABSTRACT

The study entitled “Biology and management of the two-spotted spider mite, *Tetranychus urticae* Koch (Prostigmata: Tetranychidae) on okra [*Abelmoschus esculentus* (L.) Moench]” was undertaken at Department of Agricultural Entomology, College of Horticulture, Vellanikkara during 2012-13. The objectives of the study were to elucidate the biology of *T. urticae* on okra and to evaluate selected acaropathogenic fungi viz., *Hirsutella thompsonii* and *Beauveria bassiana*, botanicals viz., neem oil 2 per cent and NSKE 5 per cent, and new acaricide molecules viz., spiromesifen 240 SC, fenazaquin 10 EC and diafenthiuron 50 WP for their bioefficacy against *T. urticae* on okra.

The study on the biology of *T. urticae* was conducted in the laboratory during October- November, 2012 at $30 \pm 2^{\circ}\text{C}$ and $61.5 \pm 7\%$ RH following leaf disc method. The life cycle of *T. urticae* consisted of egg, larva, protonymph, deutonymph and adult. The immature stages were followed by short quiescent intervals called nymphochrysalis, deutochrysalis and teleiochrysalis. The mite recorded an incubation period of 2.92 days. Larval period of 0.83 and 1.19 days, protonymphal period of 0.36 and 0.58 days and deutonymphal period of 0.67 and 0.29 days were recorded respectively in male and female *T. urticae*. The total developmental period from egg to adult emergence was shorter for male (6.73 days) compared to female (7.52 days). Adult male was smaller in size, reddish green or light red in colour and pear shaped. Adult female was broader, bright reddish in colour and globular in shape with long setae over the body and legs. Mating took place immediately after the emergence of the female. *T. urticae* exhibited both sexual and parthenogenetic reproduction. Mated female's progeny consisted of both males and females in the ratio 1:5.8, whereas unmated female produced 100 per cent males. Pre-oviposition, oviposition and post-oviposition periods lasted for 0.58, 9

and 4 days and 0.58, 11 and 4.5 days respectively in mated and unmated female. Mated and unmated females on an average produced 108 and 77 eggs. The adult mite recorded longevity of 12, 12.5 and 17 days for male, mated female and unmated female respectively. The shorter developmental period of *T. urticae* coupled with high fecundity would help the mite build up population very fast and successfully complete several generations in a crop season attaining the status of major pest.

Field studies were conducted to evaluate two acaropathogenic fungi, two botanicals and three new acaricide molecules along with a standard check and an untreated control against *T. urticae* on okra during two seasons viz., February – May, 2012 and November, 2012 – February, 2013. The new acaricide molecules diafenthiuron 50 WP @ 400g a.i. ha⁻¹ fenazaquin 10 EC @ 125g a.i. ha⁻¹ and spiromesifen 240 SC @ 100g a.i. ha⁻¹ were highly effective against *T. urticae*, bringing significant reduction in mite population. Among the botanicals evaluated, neem oil 2 per cent was found to be superior to NSKE 5 per cent during the second season though on par with each other during the first season of study. Among the acaropathogenic fungi, *B. bassiana* was found to perform better than *H. thompsonii* in terms of per cent reduction in mite and egg counts over untreated control. However consistent results are lacking for bioagents and botanicals.