EFFICACY OF *Neoseiulus longispinosus* (EVANS) (MESOSTIGMATA: PHYTOSEIIDAE) FOR THE MANAGEMENT OF *Tetranychus urticae* KOCH (PROSTIGMATA: TETRANYCHIDAE) ON CUCUMBER UNDER PROTECTED CULTIVATION

by

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(2012-21-113)

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KERALA, INDIA

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THESIS

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KERALA, INDIA

2017

DECLARATION

I, Neena Lenin hereby declare that this thesis entitled **"Efficacy of *Neoseiulus longispinosus* (Evans) (Mesostigmata: Phytoseiidae) for the management of *Tetranychus urticae* Koch (Prostigmata: Tetranychidae) on cucumber under protected cultivation"** is a bonafide record of research work 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, associateship, fellowship or other similar title, of any other University or Society.

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Certified that this thesis entitled **"Efficacy of *Neoseiulus longispinosus* (Evans) (Mesostigmata: Phytoseiidae) for the management of *Tetranychus urticae* Koch (Prostigmata: Tetranychidae) on cucumber under protected cultivation"** is a record of research work done independently by Ms. Neena Lenin under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to her.

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NEENA LENIN

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Introduction

1. INTRODUCTION

The cultivable land area in Kerala is shrinking due to high population and urbanization which resulted in a decline in food production even as the demand for food is increasing. With increase in living standards and purchasing power of people in the state, there is a growing demand for high quality food products. This necessitates production of high quality food in large quantities within the available area. The recent thrust by the state Government in intensive crop production focuses on hi- tech agricultural practices such as protected cultivation of vegetable crops, where the microclimate is modified. Polyhouse cultivation is being promoted in a big way in the state by providing 75 per cent of the cost as subsidy to the farmers. Consequently, more than 600 polyhouses have come up in Kerala for vegetable cultivation (Nambudiri, 2014).

Cucumber (*Cucumis sativus* L.) (Cucurbitaceae), is one of the most popular vegetable crops grown in polyhouses of Kerala. However, the incidence of insect pests and mites due to the favourable microclimate inside poses threat to the cultivation of cucumber in polyhouses.

Among the various pests of cucumber in polyhouse, the two spotted spider mite, *Tetranychus urticae* Koch (Tetranychidae: Prostigmata) is considered to be the most destructive. *Tetranychus urticae,* was first described by Koch in 1836 (Pritchard and Baker, 1955) is distributed globally. 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 species of plants. Farmers routinely resort to application of synthetic acaricides for mite management in polyhouses, which results in resurgence and residue problems. However, they are becoming increasingly aware of the dangers of excessive use of chemicals inside polyhouse.

Biological control of phytophagous mites using predatory mites has been proven as a successful alternative to conventional chemical control, especially in green house crops (Gerson and Weintraub, 2006). Predatory mites of the family Phytoseiidae have been the most popular so far, as they have efficiently controled mite pests in many crops around the world (Sabelis, 1981). Faunistic studies on Phytoseiidae have progressed fairly well in India and 189 species have been reported to date (Gowda and Mallik, 2010).

The phytoseiid predator, *Neoseiulus* (=*Amblyseius*) *longispinosus* (Evans) (Mesostigmata: Phytoseiidae) has been identified as one of the most potent predator of tetranychid mite, in tropics and subtropics. It has a wide distribution and has the ability to adapt to warm temperatures inside polyhouses under South Indian conditions (Mallik *et al.,* 1998). It has been reported as an important native predator of spider mites from different agricultural ecosystems across India (Arbabi and Singh, 2008; Thakur and Dinabandhoo, 2005 and Karmakar and Gupta, 2010). Binisha and Bhaskar (2013) documented the predatory mite fauna in the vegetable ecosystems of Kerala and reported *N. longispinosus* as one of the predominant predatory mite species. It was also found associated with the tetranychid mites on cucumber in Thrissur district, Kerala (Maheswary, 2015).

Biological control of spider mites using predators in polyhouses though had proven successful in several states, has not yet been exploited in Kerala. In this context, a study was conducted to evaluate the performance of *N. longispinosus* for the biological control of *T. urticae* infesting cucumber, the predominant vegetable grown under protected cultivation in Kerala. The objectives of the study entitled "Efficacy of *Neoseiulus longispinosus* (Evans) (Mesostigmata: Phytoseiidae) for the management of *Tetranychus urticae* Koch (Prostigmata: Tetranychidae) on cucumber under protected cultivation" were as follows:

- To study the incidence, crop phenology relationship and natural enemies of the two spotted spider mite *Tetranychus urticae* infesting cucumber in polyhouse
- To study the biology of *T. urticae* on cucumber
- To study the biology of *Neoseiulus longispinosus* on *T. urticae*
- To evaluate the efficacy and prey stage preference of *N. longispinosus* on *T. urticae*
- To standardize the optimum predator: prey ratio of *N. longispinosus* for biological control of *T. urticae* in polyhouse

Review of Literature

2. REVIEW OF LITERATURE

Cucumber is a major vegetable crop grown in the polyhouses of Kerala. The two spotted spider mite *Tetranychus urticae* Koch is the predominant sucking pest of cucumber in polyhouses. Currently, the mite management strategy inside polyhouse is centered on synthetic acaricides which results in adverse side effects. This necessitates the development of alternate safer strategies for the management of the two spotted spider mites on cucumber. Biological control of *T. urticae* on vegetables and ornamentals under protected cultivation using predatory mites is gaining momentum worldwide. The Phytoseiid predator, *Neoseiulus longispinosus* (Evans) has been identified as a potential candidate for mite management in polyhouse crops. Since literature pertaining to the studies on the biological control of *T. urticae* on cucumber using *N. longispinosus* is limited, similar research work conducted on other vegetable crops has also been reviewed here under.

2.1. DIVERSITY AND ABUNDANCE OF SPIDER MITES ON VEGETABLES

Spider mites in the family Tetranychidae are major pest of vegetable crops worldwide. The subfamily Tetranychinae includes a number of economically significant species, of which *T. urticae* is the most important on many vegetable crops (Nair, 1975).

Tetranychus cinnabarinus (Boisduval), *Tetranychus neocaledonicus* Andre and *Tetranychus macfarlanei* Baker and Pritchard were reported as major species of mites infesting vegetable crops of West Bengal. *T. cinnabarinus* was the predominant mite species damaging cucumber, okra, bittergourd, brinjal and beans. *T. neocaledonicus* was recorded on brinjal, bottle gourd, okra, ridge gourd, pumpkin and bitter gourd and *T. macfarlanei* on sponge gourd and pumpkin (Gupta and Gupta, 1985).

Four species of mites namely *T. cinnabarinus, T. neocaledonicus, Tetranychus ludeni* Zacher and *T. macfarlanei,* were recorded as serious pests of different vegetables *viz*., brinjal, okra, cucurbits, chilli and potato from different parts of India (Gupta, 1991).

Grewal (1992) reported *T. cinnabarinus* as the major phytophagous mite associated with brinjal in Punjab while *T. macfarlanei* (Arbabi *et al*., 1994) and *T. urticae* (Vora, 1994) were reported as the major mite species of brinjal from Varanasi and Navasari, respectively. A new species of spider mite namely *Tetranychus okinawanus* Ehara was reported as a pest from Ryuku islands of Japan on *Pueraria lobata* (Wild.) (Ehara, 1995).

Survey conducted in the vegetable fields of Thiruvananthapuram district of Kerala recorded three species of spider mites, namely, *T. cinnabarinus* on amaranthus*, T. ludeni* on okra and cowpea and *T. neocaledonicus* on cucumber, drumstick and brinjal (Sudharma, 1996). *Tetranychus urticae* and *T. cinnabarinus* were reported as serious phytophagous mites of vegetables from Punjab and Haryana (Gulati, 2004).

Tetranychus urticae was found to be a serious pest of brinjal, okra and cucumber during hot months. *Tetranychus ludeni* was found to infest cowpea and French bean, where as *T. macfarlanei* infested sponge gourd, ridge gourd, bottle gourd and pumpkin in Jharkhand (Prasad, 2006).

In Dharward, three teranychid species found infesting brinjal were *T. macfarlanei*, *T. urticae* and *Tetranychus* sp. among which *T. macfarlanei* was the dominant (Prasanna, 2007).

Tetranychus urticae was reported as the predominant phytophagous mite species associated with vegetables like pointed gourd, beans, pumpkin, cucumber, cowpea, brinjal, okra and amaranthus in West Bengal (AINPAA, 2011).

Krishna (2013) reported the occurrence of *T. urticae* on okra from Thrissur district, Kerala.

Tetranychus truncatus is a worldwide mite pest which damages cotton, corn, soybean, cucumber, kidney bean, jujube and egg plant (Guo *et al*., 2013). It is also reported as a major pest of crops in Bangladesh. Though the species is distributed across many Asian and Pacific islands (Ullah *et al*., 2014) it is considered to be a quarantine pest in Canada and US (Gotoh *et al*., 2015). Recently, *T. truncatus* was reported on cowpea, cucumber and amaranthus from Kerala while *T. macfarlanei* infested brinjal, okra, cowpea and winged bean. The study also identified *T. truncatus* as the predominant species of spider mite infesting vegetable crops of Thrissur district, Kerala (Bennur *et al.,* 2015).

2.1.1. Spider Mites of Vegetables under Protected Cultivation

Most of the mite problems in greenhouse are related to spider mites. *Tetranychus urticae* was reported as an important pest of cucurbits and other vegetables both in glass house as well as open field conditions (Meyer, 1974).

Spider mites were reported to be one of the most important pests of vegetable crops in greenhouse, affecting more than 150 plants. Significant economic losses were reported in tomato, pepper, cucumber and bean grown in greenhouses in Turkey due to spider mites (Rizzieri *et al*., 1988).

The red spider mite *T. urticae* was the predominant pest of cucumber grown under net house condition in Punjab, India (Kaur *et al*., 2010).

Tetranychus cinnabarinus was reported as a pest of carnation in polyhouses of Maharashtra and *T. neocaledonicus* as a pest of cucumber from New Delhi. *Tetranychus urticae* was the predominant species infesting tomato, capsicum, cucumber, carnation and gerbera in polyhouse (Sood, 2010).

In Mediterranean region, 93 per cent of protected cultivation is located in Turkey and the most commonly grown crops are the vegetables, namely, tomato, pepper, egg plant and cucumber. The major mite pest infesting these crops is the carmine spider mite, *T. cinnabarinus* (Yucel *et al*., 2013).

The mite species reported on cucumber under protected cultivation in Himachal Pradesh were *T. ludeni, T. neocaledonicus* and *T. urticae* (Sood *et al*., 2015).

An invasive species of tetranychid *i.e. T. okinawanus* was reported on cucumber from Thrissur district of Kerala (Bennur *et al*., 2015).

2.2. BIOLOGY OF *Tetranychus urticae*

For a better understanding of the interactions in a predator prey relationship, it is always necessary to have a thorough knowledge about the biology of the prey. The literature related to the biology of the spider mite, *T. urticae* is reviewed below.

The biology and life tables of *T. urticae* were studied on rose, bean and cucumber by Kasap (2002). The total developmental period and sex ratio were determined as 10.9 days and 0.76 on bean, 10.4 days and 0.65 on cucumber and 11.2 days and 0.68 on rose, respectively. The study also pointed out that bean and cucumber were more preferred hosts than rose.

Riahi *et al*. (2013) conducted a study on the influence of temperature on development and reproduction of *T. urticae* with in a temperature regime of 13 ºC to 33 ºC. The results showed that the developmental time increased at higher temperatures (27 ºC to 33 ºC) and decreased at lower temperatures (17 ºC to 27 ºC). No mite development was observed at 13 ºC.

In a study conducted by Rajkumar (2003), the total life cycle of *T. urticae* on jasmine were 10.70 days for males and 12.36 days for females. The egg, larva, protonymph, deutonymph, oviposition and longevity of females lasted for 4.46, 2.72,

2.33, 1.50, 14.5 and 18.7 days, respectively. The developmental periods of males were 4.30, 2.42, 1.66 and 1.30 days for egg, larva, protonymph and deutonymph, respectively with longevity of 12.1 days.

Under favourable temperature, spider mites could complete their life cycle in as little as 5 days (Fahnbulleh, 2007).

Hoque *et al*. (2008) studied the influence of seasons on the biology and life tables of *T. urticae* under laboratory conditions during summer, winter and autumn. The total developmental period during summer, autumn and winter varied from 7- 8, 9- 10 and 17- 19 days, respectively. In all the three seasons, the male to female ratio of 1: 2.9 remained the constant.

The influence of temperature on the developmental stages of *T. urticae* was studied at room temperature in different months. The study by Naher *et al*. (2008) revealed that the duration of development was shortened (4.22 days) during hot months and the developmental period was extended upto 28.33 days during winter period.

Life history studies of *T. urticae* on gerbera by Silva *et al*. (2009) recorded the development of egg, larva, nymphochrysalis, protonymph, deutochrysalis, deutonymph and teliochrysalis in 3.15, 3.46, 1.13, 2.10, 1.07, 1.63 and 1.11 days, respectively. The viability of eggs of unmated and mated females was found to be 96.5 per cent and 97.1 per cent, respectively.

Razmjou *et al*. (2009) conducted studies in Iran on the influence of host plants on the developmental biology of *T. urticae* on three legume cultivars. The fecundity and longevity of *T. urticae* showed significant variation among the cultivars where as the egg hatchability and developmental time were found to be similar.

In a study conducted on pear varieties in Iran, by Riahi *et al*. (2011) it was found that the total developmental period of *T. urticae* from egg to adult varied

among the three pear varieties. But the pre- oviposition, oviposition, post- oviposition periods, fecundity and longevity did not differ significantly among the varieties.

El- Wahed and El- Halawany (2012) observed a negative correlation between the temperature and incubation period of spider mites. The longest incubation period of female was 13.6 days at 15 ºC and shortest was 2.7 days at 30 ºC. Though the duration of life stages and life span was reduced, rate of reproduction increased along with temperature. The highest mean fecundity (156. 8 eggs) was observed at 30 ºC.

Studies on the biology of two spotted spider mite, *T. urticae* in okra by Krishna and Bhaskar (2014) revealed that the incubation period was 2.92 days. The larval, protonymphal and deutonymphal periods of male were 0.83, 0.36 and 0.67 days and that of female were 1.19, 0.58 and 0.29 days, respectively. The mite exhibited parthenogenetic as well as sexual reproduction. Unmated females produced only males. The unmated females lived longer (17 days) than mated ones (12.5 days). Sex ratio of the progenies of the mated females was 1:5.8.

2.3. NATURAL ENEMIES OF *Tetranychus urticae* ON VEGETABLES

An array of natural enemies have been recorded against the two spotted spider mite. The most important ones are discussed here.

2.3.1. Insect Predators of Spider Mites

Several insects belonging to the families Anthochoridae, Cecidomyiidae, Chrysopidae, Coccinellidae, Staphylinidae and Thripidae prey upon phytophagous mites.

The major species of coccinellids reported as specific predators of mites are *Stethorus pauperculus* Weise (Kapur, 1948), *S. tetranychi* Kapur (Puttarudriah and Channabasavanna, 1955), *S. keralicus* Kapur (Puttaswamy and Rangaswamy, 1976), *Parastethorus histrio* (Chazeau) (Dhooria, 1981), *S. parcepunctatus* Kapur (Gupta, 2001) and *S. gilvifrons* (Mulsant) (Sarmah and Bhattacharya, 2002).

Several predators like *Stethorus* spp., *Oligota* spp., *Anthrocnodax occidentalis* Felt, *Feltiella minuta* Felt, *etc.* were reported on spider mites in Taiwan. Green lacewings (*Mallada basalis* Walker and *Chrysoperla carnea* Stephens) were also effective generalist predators of spider mites (Ho *et al.,* 1995).

Abhilash (2001) reported the green lace wing, *Chrysoperla carnea* (Stephens) as a potential predator of *T. ludeni* on cowpea. Several species of coccinellids have been reported as general predators of mites namely, *Brumoides suturalis* (F.), *Cheilomenus sexmaculata* (F.), *Coelophora bissellata* Mulsant, *Hippodamia variegata* (Goeze), *Micraspis univittata* (Hope), *Cryptogonus quadriguttatus* (Weise), *Scymnus coccivora* Ayyar, *S. xerampelinus* Mulsant, *S. nubilus* Mulsant, *Jauravia soror* (Weise) and *Pharoscymnus flexibilis* (Mulsant) (Omkar and Pervez, 2004).

The predators, *S. gilvifrons* and *Scolothrips longicornis* Priesner were found to play a major role in the natural bio control of the two spotted spider mite on castor bean trees in Egypt (Ismail *et al*., 2007).

An investigation was carried out on the natural enemies of red mite, *T. bioculatus* in the marigold gardens by Taleb and Sardar (2007). Two species of lady bird beetles *viz., Stethorus punctillum, Micraspis discolor* and two species of spiders *viz.,* lynx spider (*Oxyopes* sp.) and long mouth spider (*Tetragatha* sp.) were found to be predatory on the mite. The abundance of predator population was positively correlated with the abundance of their mite prey.

Insect predators like *Oligota* sp., *Scolothrips rhagebianus* Preiesner and two unidentified predatory bugs belonging to the family Anthocoridae and Miridae were observed to feed on tetranychid mites on brinjal. *Geocoris* sp. was found to be a facultative predator of spider mites (Prasanna and Kumar, 2008).

Studies on the evaluation of an anthocorid predator *Blaptostethus pallescens* Poppius against two spotted spider mites by Ballal *et al.* (2009) proved that the predator could bring about 78 per cent reduction in the mite population on okra.

Members of the subfamily Stethorini of the family Coccinellidae are principally feeders of tetranychid mites. Earlier studies indicated that Stethorini are high density predators and are unable to regulate spider mite populations at low densities (Biddinger *et. al*., 2009).

Perumalsamy *et al.* (2010) reported the predatory beetles, *Oligota pygmaea* (Solier), *O. oviformis* Casey and *S. gilvifrons* on *Olygonychus coffeae* (Nietner) in tea.

A laboratory study was carried out in Tamil Nadu to find out the influence of prey densities on the efficacy of *S. pauperculus* and *Oligota* sp. The predator prey ratio of 5:150 was found to be the ideal for both the beetle predators (Jeyarani *et al*., 2012).

In a study conducted at Punjab Agricultural University, Ludhiana, *Chrysoperla zastrowi arabica*, *Chrysoperla carnea* and *Blaptostethus pallescens* Poppius were found to be potential predators of *T. urticae* (AINPAA, 2011).

2.3.2. Fungal Pathogens of Mites

The first record of a *Neozygites* species infecting spider mites was made by Fisher in 1951, who observed adult mortality following infection. Weiser and Muma (1966) described *Neozygites floridana* Fisher as a pathogen of the Texas citrus mite, *Eutetranychus banksi* (MacGregor) while Weiser (1968) described *Neozygites tetranychi* (Weiser) on the two-spotted spider mite, *T. urticae* from fruit orchard of Czech Republic.

Hirsutella sp. was first reported as a pathogen of eriophyid mite and citrus rust mite from Florida back in 1924 by Speare and Yothers. *Hirsutella thompsonii* was observed to cause mortality in *T. cinnabarinnus, Eutetranychus orientalis* and *Panonychus citri* (McCoy, 1981).

Paecilomyces eriophytis, *P. farinosus*, *P. fumosoroseus* and *P. terricola* (Chandler *et al*., 2000) and a species of *Sporothrix* (Kumar *et al*., 2004) were also known to infect spider mites.

Natural isolate of *Beauveria bassiana* (Balsamo) was reported to be more virulent than other cultures of *B. bassiana* against *T. urticae* (Ghosh *et al*., 2007). Natural infection of *B. bassiana* on tetranychid mites was reported on tomato, beans and okra from Karnataka during 2004- 2005. The infection rate was 12. 94 per cent on beans (Kalmath *et al*., 2007).

Cladosporium cladosporoides (Frensen.) recorded 75 to 95 per cent natural infection on spider mites on cowpea and okra crops in Coimbatore (AINPAA, 2011)

Large scale mycosis of *T. urticae* was reported from Kerala on spider mite culture maintained in polyhouses on brinjal. Three fungal pathogens *viz*., *N. floridana*, *Acremonium zeylanicum* (Petch) W. Gams and *Conidiobolus* sp. were isolated from these mycosed mites. Natural epizootic of *A. zeylanicum* on mite was the first record for India (Krishna *et al*., 2014).

2.3.3. Predatory Mites of Spider Mites

Gupta and Gupta (1985) reported 10 species of predatory mites on vegetables namely *Amblyseius largoensis* (Muma), *A. ovalis* (Evans), *A. alstoniae* Gupta, *A. longispinosus* (Evans), *Cunaxa* sp., *C. setirostris* (Hermann), *Agistemus* sp., *Spinibdella* sp., *Rhizoglyphus* sp. and *Tyrophagus putrescentiae* (Shrank) on different vegetables in West Bengal.

Gupta (1991) recorded 17 species of mite predators from major parts of India, of which five species *viz*., *Amblyseius multidentatus* Swirski and Shechter, *Amblyseius alstoniae* Gupta, *A. delhiensis* (Narayanan and Kaur), *A. finlandicus* (Oudemans) and *A. tetranychivorus* Gupta were the major ones. The major predatory mites associated with brinjal in Punjab were *A. alstoniae, A. finlandicus, A.*

multidentus, Phytoseius roseus Gupta, *P. minutus* Narayanan and Ghai, *Typhlodromus gopali* Gupta, *Agistemus industani* and *Pronematus* sp. (Grewal, 1992).

A field study in Gujarat revealed the presence of the predatory mites *Phytoseius* sp. and *Pronematus flaschneri* in association with the phytophagous mites of brinjal (Vora, 1994).

Abhilash (2001) reported the predatory mite, *Amblyseius* sp. as potential predator of *T. ludeni* on cowpea. Zacarias and Moraes (2002) reported about 31, 603 mites under 105 species from Brazil among which 43 species belonged to predatory families.

A survey on predatory mite fauna in Kozhikode and Malappuram districts of Kerala revealed 40 species under nine genera, of which two were new reports *viz*., *Amblyseius bhadrakshae* (Sadanandan and Ramani) and *A. amorphalae* (Sadanandan and Ramani) (Sadanandan and Ramani, 2006).

Amblyseius longispinosus and *P. minutus* were the prevalent predatory species associated with the tetranychids in brinjal ecosystem in Dharwad (Prasanna, 2007). The only predatory mite found predating on tetranychid mites in brinjal was *A. longispinosus* (Prasanna and Kumar, 2008).

Of the 31 species of predatory mites observed by Karmakar and Gupta (2010), four species *viz.*, *A. longispinosus, A. largoensis* and *Agistemus* sp. were reported as dominant and efficient predators. In a survey conducted to record the phytoseiid mites of Southern Karnataka, Gowda and Mallik (2010) reported fifty one species of phytoseiid mites of which 21 were new to science.

A survey on the phytoseiid mites and associated pests was carried out in two districts of Kerala, Kozhikode and Malappuram. Among the 18 species of phytoseiid mites identified, five species *viz., A. largoensis*, *Amblyseius suknaensis* Gupta, *A. guptai, Paraphytoseius multidentatus*, and *Phytoseius rachelae S*wirski and Shechter were recognized as the most common and abundant phytoseiid predators in the

region. Of these, *A. largoensis*, *T. suknaensis* and *A. guptai* had broad range of feeding habits. On the other hand, *P. multidentatus* and *P. rachelae* showed more preference to insect pests than mite pests (Sheeja, 2010).

The predatory mites associated with the phytophagous mites of egg plant and cucumber in Turkey were *Phytoseius finitimus* Ribaga and *Amblyseius andersoni* (Chant), respectively (Ozisli and Cobanoglu, 2011).

A study was carried out by Ohno *et al.* (2012) to identify the available phytoseiid mites of Okinawa Island, Japan. Out of the19 species recorded, *Neoseiulus womersleyi* (Schicha) was the predominant one, followed by *Amblyseius eharai* Amitai and Swirski.

Study on the relative abundance of predatory mites associated with phytophagous mites in different vegetable crops of Kerala revealed the occurrence of 15 species of mites belonging to six genera *viz*., *Amblyseius*, *Typhlodromips*, *Neoseiulus*, *Euseius*, *Phytoseius* and *Paraphytoseius* under the suborder Mesostigmata. In the genus *Amblyseius,* eight species were recorded *viz*., *A. aerialis, A. indirae, A. channabasavannai, A. kundurukkae, A. largoensis, A. orientalis, A. herbicolus* and *A. kulini.* Two species recorded in the genus *Euseius* were, *E. coccinea* and *E. alstoniae*. *Typhlodromips syzygii* was the only species recovered from the genus *Typhlodromips.* The genus *Phytoseius* included the species, *P. wainsteini* and *P. punjabensis*. A single species was obtained from genus *Neoseiulus, viz*., *N. longispinosis. Paraphytoseius scleroticus* was the only species in the genus *Paraphytoseius.* The relative abundance of the various genera recovered during the survey was represented as *Amblyseius* > *Euseius* = *Phytoseius* > *Neoseiulus* =*Typhlodromips* = *Paraphytoseius* (Haneef and Sadanandan, 2013).

The major predatory mites associated with vegetables in Thrissur district of Kerala belonged to five families, of which the members of the family Phytoseiidae were the dominant. *A. paraaerialis* Muma, *P. orientalis* Narayanan, Kaur and Ghai, *N. longispinosus*, Phytoseius sp., *Euseius macrospatulatus* Gupta, *Typhlodromips* sp.

and *Scapulaseius* sp. were the Phytoseiids recorded. Other predatory mites observed were *Tydeus* sp., *Cunaxa* sp., *Bdella* sp. and *Agistemus* sp. (Binisha and Bhaskar, 2013).

A survey was undertaken to explore the predatory mite fauna associated with the vegetable crops of Thrissur district of Kerala *viz.,* amarathus, brinjal, okra, bittergourd, chilli, cowpea, coccinia, cucumber, snakegourd and snap melon. A total of 17 predatory mite species belonging to five families *viz.,* Phytoseiidae, Stigmaeidae, Cunaxidae, Bdellidae and Tydeidae represented by 12 genera were encountered. Brinjal recorded the highest species richness of predatory mites with 17 species. Phytoseiidae was found to be the predominant mite family and the major species recorded were *N. longispinosus, A. paraaerialis, A. largoensis, E. macrospatulatus, Euseius* sp. nr. *prasadi, T. syzygii, P. orientalis, Phytoseius intermedius* and *Scapulaseius sp.* Other major predatory mites recorded were *Cunaxa* sp. (Cunaxidae), *Bdella khasyana* (Bdellidae), four species of the family Stigmaeidae *viz., Agistemus gamblei, A. fleschneri, A. garrulus* and *A. macrommatus* and two species of Tydeidae *viz., Tydeus gossabaensis* and *Pronematus anconai. Euseius* sp. nr. *prasadi, P. intermedius*, *B. khasyana*, *A. fleschneri, A. garrulus, A. macrommatus*, *T. gossabaensis* and *P. anconai* were reported for the first time from Kerala (Maheswary, 2015).

2.4. BIOLOGICAL CONTROL OF SPIDER MITES USING PHYTOSEIID MITES

Predatory mites of the family Phytoseiidae are of economic importance because they efficiently control pest mites in many crops around the world (Sabelis, 1981). Phytoseiid mites have received global attention since the 1950's due to their importance as natural predators of phytophagous mites and small insects and consequent usefulness in the biological control of crop pests (Swirskii and Amitai, 1997). The literature related to the biology, mass culturing, predatory potential and predator prey ratios of phytoseiids with special reference to *N. longispinosus* is discussed here under.

2.4.1. Biology of Phytoseiid Predators

Laing (1968) studied the life history of *Phytoseiulus persimilis* on *T. urticae* in growth chambers in which the temperature varied from 18- 35°C. The study revealed that the mean developmental period of *P. persimilis* from egg to adult was 7.45 days. The mean generation time was 17.32 days; the maximum rate of increase for the predator was higher than that for the prey. The longevity of the female predator was around 50 days.

Kazak *et al.* (2002) studied the development time, survival and fecundity of the generalist predatory mite, *Neoseiulus umbraticus* Chant at 20°C, 25°C, and 30°C and 65 ± 10 per cent RH. *Neoseiulus umbraticus* females completed development in 9.7, 8.0 and 5.9 days, respectively, using a diet of all life stages of *T. cinnabarinus*. Total developmental period for male was relatively shorter at 25 °C and 30°C than at 20°C. In general, pre- oviposition, oviposition, and post- oviposition periods of *N. umbraticus* shortened as temperature increased. The longest survival of *N. umbraticus* of 80.5 days occurred at 20°C, followed by 67.0 and 57.6 days at 25°C and 30°C, respectively. Mated females laid an average of 0.9, 1.3 and 1.4 eggs per female per day and 33.1, 44.0 and 43.6 eggs over their entire lives at 20°C, 25°C and 30°C, respectively.

Neoseiulus baraki developed and multiplied satisfactorily on the storage mite, *T. putrescentiae*. The predatory mite, when fed on *T. putrescentiae* developed from egg to adult in 11.1 ± 0.1 days and deposited 26.4 ± 2.2 eggs during a life-time of 70.0 ±1.8 days. It multiplied equally well on pollen of *Typha* sp. and rice bran. The fecundity of *T. putrescentiae* was 163.5 ± 23.4 eggs on *Typha* pollen and 143.7 ± 9.7 eggs on rice bran. But the daily oviposition rate was higher on rice bran (Fernando *et al*., 2004).

Goshal *et al*. (2004) studied the biology of the predatory mite *A. multidentatus* with *Eotetranychus fremonti* Tuttle and Baker as prey. The incubation, larval,

protonymphal and deutonymphal period were 1.72, 1.56, 1.60 and 2.30 days, respectively. The total development period from egg to adult was completed in 7.35 days. The pre- oviposition, oviposition and post- oviposition periods were respectively 1.43, 14.52 and 6.62 days. The adult lived for 12.01 days. A female on an average laid around 18.01 eggs and 79.56 per cent of the eggs hatched out to larva. The sex ratio was female biased and was recorded as 1: 3.5.

2.4.1.1. Biology of Neoseiulus longispinosus

Ganok (1982) reported that longevity of females of *A.* (=*Neoseiulus*) *longispinosus* was much higher than that of *T. truncatus* females.

Studies conducted at the Entomology Laboratory, Bogor Agriculture Institute revealed that the life cycle of *A. longispinosus* was completed in 4.78 days. The female recorded longevity of 15.42 days and fecundity of 25.90 eggs. Reproduction value for all ages was 56.66. Stable age distributions at laboratory constituted 42.42 per cent eggs, 31.94 per cent nymphs and 25.94 per cent adults (Puspitarini, 2010).

The life history and demography of *A. longispinosus* on the two spotted spider mite, *T. urticae* was studied under laboratory conditions of 25- 28°C and 65- 85 per cent RH. The life cycle was completed in 102. 5 h for males and females with 90 per cent survival. The incubation period of male was 45.2 h which was slightly longer than that of females (42.6 h). The larval, protonymphal and deutonymphal periods for both the sexes were 15.7, 21.1 and 23.0 h, respectively. Mating occurred on the same day of adult emergence and was repeated several times during the reproductive life. Egg laying started on the second day after emergence. Parameters relating to oviposition were: fecundity, 50.7eggs/female; hatchability, 99.6 per cent and oviposition period, 28 days. Net reproductive rate was 36.7 female offspring/female/generation time of 9.0 days. The sex ratio was biased toward the females (71. 9 per cent) while the intrinsic rate of increase was 0.4 with a doubling time (DT) of 1.7 days. Males lived longer (36 days) than females (30 days) with a LT~ 0 of 26 days for males and 22 days for females (Ibrahim and Palacio, 1994).

The biology of *A. longispinosus* was studied on *Eotetranychus cendanai* Rimando. Egg, larval, protonymphal and deutonymphal stages lasted for 2.0, 0.57, 1.07 and 1.16 days, respectively for males and 0.13, 0.17, 0.31, 0.53 days, respectively for females. The egg to adult period was 4.79 days. Females survived for 14.61 days and the fecundity was 19.54 eggs/ female. A single female laid 1.33 eggs/ day (Thongtab *et al*., 2001).

Kongjarean (2006) studied the developmental history of *N. longispinosus* using *P. latus* as prey. The egg, larval, protonymphal and deutonymphal period was found to be 1.25 ± 0.26, 0.30 ± 0.10, 0.90 ± 0.20 and 0.65 ± 0.13 days, respectively. The adult male lived for 6.55 ± 0.51 days and females for 8.5 ± 0.68 days.

Neoseiulus longispinosus, is a potential predator of the polyphagous spider mite, *Tetranychus kanzawai* Kishida. The life cycle of *N. longispinosus* was short, *viz.* 3.23 days. The oviposition period was 8.76 days with maximum daily egg production of 4 eggs/female/day when the predatory mite was six days old. Fecundity of *N. longispinosus* increased with the prey population; so the numerical response of *N. longispinosus* was direct response. *N. longispinosus* preferred to consume eggs than the other stages of *T. kanzawai* (Song *et al*., 2016).

Madruga *et al.* (2012) studied the developmental duration, mortality, sex ratio, reproduction and survival of *N. longispinosus* feeding on *Tetranychus tumidus* Banks. Laboratory conditions of 34 ± 2. 9°C temperature and 73.54 ± 1.04 per cent relative humidity were set for the study. The developmental period from egg to adult was found to be seven days. The mean pre- oviposition, oviposition and longevity of females were 2.3, 11.5 and 28 days, respectively. The total fecundity was 21.70 ± 9.11 eggs. The sex ratio was 0.70 and was female biased.

Total life cycle of *N. longispinosus* was found to range between 7 to 14 days in the winter months of November and December (Sharma and Chauhan, 2013).

2.4.2. Mass Culturing of *Neoseiulus longispinosus*

Although biological control of spider mites using predatory phytoseiid mites is feasible (McMurtry and Croft, 1997) mass rearing of predatory mites for the purpose is a complex affair as it requires the maintenance of stable tri- trophic systems comprising of the predators, their prey and the host plants of the prey.

For utilizing *N. longispinosus* in biological control of spider mites, it should be mass reared on host plants suitable for rearing the prey mites in greenhouses or in the fields (Hoy *et al.,* 1982). Further, the optimum ratio of predator: prey for release also needs to be decided in order to harvest the maximum number of predators as quickly as possible (Overmeer, 1985). *Neoseiulus longispinosus* could be easily mass reared on detached mulberry leaves with the mulberry red mite, *T. truncatus* as prey (Kongchuensin *et al.,* 1989).

Hegde and Patil (1995) reported the mass multiplication of the predator, *A. longispinosus* on potted cotton (MCV- 5) plant harbouring cotton red spider mite, *T. macfarlanei.* Gravid females of *A. longispinosus* were released at densities of 1, 2, 3, 4, and 5 pairs per plant and count was made 10 days after the release of predator. A total number of 4, 7, 12, 15 and 20 predators were recorded from initial 1, 2, 3, 4 and 5 pairs per plant, respectively.

Vaidya (1999) as well as Mallik *et al.* (1999) studied the mass production of *A. longispinosus* on potted plants in a glass house at 40- 41°C and 30 per cent RH. Twelve days after sowing, when the seedlings attained nine leaflets stage, spider mites were released, followed by release of predatory mites nine days later. Four thousand seven hundred and fifteen predators were harvested from 15 French bean plants at a cost of production of rupees 0.02 per predator.

DeLeon and Corpuz (2005) evaluated cheap alternative preys for mass rearing of *A. longispinosus*. Survival, consumption and reproduction of *A. longispinosus* on eggs of different preys were evaluated singly or in combination with castor pollen as food supplement. The predator failed to develop and reproduce normally when fed on

castor pollen alone. Only the natural prey spider mites, *T. truncatus* and *T. kanzawai* (Kishida), were suitable for the predator.

Kongchuensin *et al.* (2006) reported that *N. longispinosus* could be mass-reared on mung bean, cowpea and soybean plants with *T. truncatus* as prey. The adult females per leaflet of mung bean and cowpea were more compared to soybean. The study also revealed that the best harvesting time of *N. longispinosus* would be two weeks after inoculation. Three weeks after inoculation, the physiological condition of the bean plants seriously deteriorated due to heavy mite injury, and as a result, the prey abandoned the host plants, resulting in a reduction in the numbers of the predator.

To determine the optimum initial ratio of predator: prey for attaining the best harvest of *N. longispinosus*, laboratory studies were conducted using *T. truncatus* on cowpea plants in greenhouse. Six different initial release ratios of predator: prey was examined for mass rearing of *N. longispinosus*. One week after inoculation, the number of *N. longispinosus* was significantly higher ($p<0.05$) at the 1: 10 ratio than in other ratios. Two weeks after inoculation, the number of predators greatly increased. The number of predators was significantly higher at 1: 20, 1: 30, or 1: 40 ratios than that at 1: 10 or 1: 60. Three weeks after inoculation, the number of *N. longispinosus* decreased to very low levels for all ratios, and there was no significant difference between the ratios. The results concluded that the optimum initial predator: prey ratio to obtain the largest number of *N. longispinosus* would be 1: 20- 1: 40 (Kongchuensin *et. al*., 2006).

Neoseiulus longispinosus is an obligatory predator of spider mites that can be mass multiplied only on spider mites. The suitable host plant for mass multiplication was identified as French bean. Release of ten prey mites per leaflet on 20 days old French bean plants, followed by the release of two or four predators per leaflet yielded one thousand predators in ten days (Jayasinghe, 2008).

Sharma and Chauhan (2013) standardized techniques for the mass multiplication of *N. longispinosus* using potted bean plants. Two pairs of phytophagous mites were released on bean seedlings at two leaf stage. Once the spider mites established, gravid females of the predator was released.

2.4.3. Predatory Potential of *Neoseiulus longispinosus*

The predatory mite *A.* (= *Neoseuilus*) *longispinosus* was explored as a potential biocontrol agent against the spider mite, *Aponychus corpuzae* in China by Zhang *et al.* (1998). The rate of prey consumption increased linearly with temperature from 1.35 at 15°C to 5.22 at 35°C, whereas the rate of oviposition increased with temperature from 0.11 at 15°C to 3.58 at 30°C and then decreased to 2.27 at 35°C. The rate of predation increased linearly with prey density within the range of 1-9 prey per leaf and reached a plateau at the density of 9 prey per leaf, but the rate of oviposition increased linearly with prey density. The number of eggs laid by predators increased linearly with the number of prey they consumed. On an average, a predator consumed 1.3- 2.5 prey to lay 1.0- 1.5 eggs. Functional responses of predator to prey density at five different temperatures (15, 20, 25, 30 and 35°C) and to different active life stages of spider mites (larva, protonymph, deutonymph, adult female and adult male) approximated Holling type II. *Amblyseius longispinosus* was considered more effective against spider mites at 25°C than at other temperatures. Similarly, they are more effective against adult female spider mites compared to nymphs. With a fixed number of prey available, predation rates decreased with predator density. At high prey densities, *A. corpuzae* adults were observed to show defensive behaviour against predators.

The predatory potential of *A. longispinosus* was explored against *Schizotetranychus nanjingensis* Ma & Yuan. Functional response experiments at six different temperatures showed that handling time (*Th*) generally decreased with temperature, whereas successful attack rate (*a*) increased with temperature and leveled

off at >20ºC. Judged by *a/Th* values, *A. longispinosus* was most efficient against *S. nanjingensis* at 30-35ºC, about half as efficient at 20 and 25ºC and performed poorly at 10-15ºC. The rate of oviposition increased linearly with prey density. As expected, the number of eggs laid by predators increased linearly with the number of prey they consumed. With a fixed number of prey available, predation rates decreased with predator density (Zhang *et al.*, 1999).

Ibrahim and Rahman (1997) investigated the influence of prey density and developmental stages on the predatory behaviour of *A. longispinosus,* using *T. urticae* as prey. Gravid females were observed to be more voracious compared to the immature female. The feeding potential showed an increasing trend with the increasing density and leveled off at a prey density of 40 per predator. The highest mean number of eggs consumed in 24 h was 16.70 for the young female and 33.30 for the gravid female. The highest mean number of larvae consumed in 24 h by the young female was 17.00 and 27.80 for the gravid female. The predator reached a satiation point at a lower density of five to ten adult prey per female predator. In general, the response curves were adequately described by the Holling's Type II model. Under continuous exposure for five days, a significant reduction in consumption was observed with the gravid female from the second day onwards, to a level similar to the number of eggs and larvae consumed by a young female predator.

Thongtab *et al.* (2001) reported that *A. longispinosus* could be employed as the biological control agent of *E. cendanai* both in laboratory and greenhouse conditions at the predator: prey ratio of 1: 30 to 1: 50.

A study on the efficacy of *N. longispinosus* for controlling *P. latus* in laboratory as well as in greenhouse was carried out at predator prey ratios of 1: 100, 1: 50, 1: 25, 1: 10 and a control. The results showed that the predator prey ratio of 1: 10 could be employed to control the mite (Kongjarean, 2006).

Surveys conducted in tea gardens of The Anamallais (Tamil Nadu, India) revealed the presence of a predatory mite, *N. longispinosus* which feeds on all stages of *O. coffeae*. A study was carried out in South India to determine the predatory potential, prey stage preference and optimum predator: prey ratio of *N. longispinosus* under laboratory and green house conditions using *O. coffeae* as prey mite. After eight days of release, the number of adult *O. coffeae* decreased and the number of predators increased. *N. longispinosus* was found feeding on all life stages of *O. coffeae* (Rahman *et al*., 2012 a).

2.4.4. Prey Stage Preference

Several studies have confirmed that *N. longispinosus* do ensure preference between different life stages of the prey.

In a study conducted by Mallik *et al.* (1998), *N. longispinosus* consumed more eggs than nymphs and adults of *T. urticae* on rose.

The predatory mite, *N. longispinosus* showed preference towards younger stages (larva and nymph) of *O. coffeae* than eggs (Rahman *et al*., 2012 a).

A study was undertaken at Gandhi Krishi Vigyan Kendra, Bangaluru, to ascertain the efficacy of *N. longispinosus*, as a biological control agent for managing two spotted spider mite on tomato in polyhouses. The results showed that at a ratio of 50:1, predators required 5 weeks to eliminate the spider mites completely from the tomato plants, while at release ratio of 100: 1, predator eliminated the spider mites in 6 weeks. At ratios of 200:1 and 400:1 spider mites were not eliminated completely during the study period. Lowest proportion of eggs was found at the ratio 50:1 (0.74 eggs/leaflet) followed by 200:1 (15.98), indicating predators prefer eggs than other stages of the spider mite (Jayasinghe and Mallik, 2014).

2.4.5. Optimum Predator: Prey Ratio of *Neoseiulus longispinosus* for Biological Control of Spider Mites

Hsiao (1988) reported that releasing *N. longispinosus* at the rate of 1:5, 1:10 or 1:20 was effective in controlling the population of *T. kanzawai* in tea plantations.

Manjunatha (1988) studied the interaction between *A. longispinosus* and *Oligonychus indicus* at five ratios (1: 1 0, 1: 20, 1: 30, 1: 40 and 1: 50). Irrespective of the ratios, the predator population assumed proportion sufficient to counter the increasing prey population. Prey elimination from milieu was on 12^{th}, 18^{th}, 20^{th}, 24^{th} and 30^{th} day at the above ratios, respectively. The decline in proportion of the prey corresponded with the appearance of the nymphs of the predators.

Anil (1990) studied the interaction between *A. longispinosus* and *O. indicus* at ratios of 1: 5, 1: 10, 1: 20 and 1: 30. He observed that irrespective of the ratios, the predator population assumed proportions sufficient to counter the increasing prey population. Prey elimination from the milieu was on 11^{th} and 13^{th} day at the above ratios, respectively.

Mallik *et al.* (1998) reported that *A. longispinosus* caused maximum reduction of tetranychid *(T. urticae)* population when they were released at a ratio of 1: 300 compared to 1: 450 and 1: 900 on rose plants. Reduction in number of tetranychid eggs was higher compared to that of nymphs and adults. Good control of tetranychid mites was achieved in 21 days after release of predators. Predators spread to the predator free plants when the prey mites were exhausted on the released plants. The results of the study indicated that, when released at 1: 100 ratio, the prey was suppressed in about seven to fourteen days.

Amblyseius longispinosus was found to be effective against *T. urticae* population when released at a ratio of 1: 300 compared to 1: 450 and 1: 900 on rose plants (Onkarappa, 1999).

Kongchuensin *et al.* (2001) studied the effectiveness of *A. longispinosus* on *T. urticae* in a strawberry field in Thailand, at a rate of two, five and 10 predators per plant when *T. urticae* number was five per leaflet. Spider mite population was significantly reduced within four weeks after releasing the predator two times. Mass releases were done at two weeks interval at the rate of two to five predators per plant seven times. The results showed that spider mite population was 172.64 mites per leaflet in the check, but 57.86 mites per leaflet in the predator released plot.

It was found that *N. longispinosus* could eliminate *T. urticae* completely, five weeks after release at a ratio of 1: 50 inside polyhouse (Jayasinghe, 2008).

Potential of *N. longispinosus* to control spider mites on rose and carnation in polyhouses has been assessed. Studies showed that at a predator: prey ratio of 1:50, the time taken to reduce prey population was less compared to wider ratios like 1: 200. When the predator prey ratio reached 1: 23, *N. longispinosus* abandoned the patch and hence the effective and economical control of spider mites could be achieved by using a ratio lower than 1: 25 (Rajashekharappa, 2010).

The optimum ratio of predators to prey for the control of red spider mite population by *N. longispinosus* was determined in laboratory. The results revealed that the predator prey ratio of 1: 33 and 1: 50 were effective. Evaluation inside greenhouse condition identified 1: 25 as the best predator prey ratio for augmentative release (Rahman *et al*., 2012a).

The research findings of a study carried out at Tamil Nadu Agricultural University, Coimbatore revealed that release of *N. longipinosus* at the rate of 20 and 25 numbers per plant could effectively control the spider mite population and was at par with the acaricide, Propargite 2.5 ml/l (AINPAA, 2013). The predator: prey ratio required to wipe out population of *T. urticae* in a week was found to be 1: 30 during July- August and 1: 17 during November- December (Sharma and Chauhan, 2013).

Materials & Methods

3. MATERIALS AND METHODS

The present study was undertaken at the Acarology laboratory, Department of Agricultural Entomology, College of Horticulture, Vellanikkara during 2013-2016. The objectives of the investigation were to study the incidence, crop phenology relationship and natural enemies of the two spotted spider mite *Tetranychus urticae* Koch infesting cucumber in polyhouse, the biology of *T. urticae* on cucumber, biology of *Neoseiulus longispinosus* (Evans) on *T. urticae*, evaluation of the efficacy and prey stage preference of *N. longispinosus* on *T. urticae* and standardization of optimum predator: prey ratio of *N. longispinosus* for biological control of *T. urticae* in polyhouse.

The method and procedures adopted for conducting various experiments based on the objectives are presented in this chapter.

3.1. INCIDENCE, CROP PHENOLOGY RELATIONSHIP AND NATURAL ENEMIES OF *Tetranychus urticae*

Periodical surveys were conducted to study the incidence, diversity, crop phenology relationship and natural enemies of *T. urticae* infesting cucumber in polyhouse.

3.1.1. Diversity of Phytophagous Mites on Cucumber in Polyhouse

Purposive surveys were conducted from June 2013 to June 2015 in 15 randomly selected polyhouses (Table 1) in four districts of Kerala, namely, Palakkad, Wayanad, Thiruvananthapuram and Thrissur (Plate 1 and 2). Mite infested cucumber leaves were randomly collected in polythene bags from each polyhouse and were brought to the laboratory. The mite specimens were then randomly picked from the infested leaves for slide mounting using Hoyer's media. Adult male and female mites from each colony were mounted separately. The males were mounted in the lateral position for better orientation of aedeagus, which is the key character for

Table 1. Polyhouses surveyed during June 2013 to June 2015

District	Locality	Owner	Period
Thrissur	Anthikkad (10.4097°N; 76.1262° E)	Mr. Suresh Babu	Aug – Dec, 2013 Jan – June, 2014 Nov – Dec, 2014 March, 2015
		Mr. Rajan Vallath	Aug – Oct, 2013
		Mr. Mohammad	Dec, 2014
		Mr. Sugathan	Dec, 2014
		M/s Elite	Aug, 2013 Feb –April, 2014
	Vellanikkara (10.5452° N; 76.2740° E)	Dept. of Olericulture	June – Oct, 2013
	Chavakkad (10.5833° N; 76.0189° E)	Mrs. Susheela Krishnakumar	Oct – Dec, 2013 Jan – May, 2014
		Mrs. Nandhini	Oct – Dec, 2013
	Kunnamkulam (10.6516° N; 76.0711° E)	Mr. Sanal Kumar	March, 2015
Palakkad	Elavanchery (10.5957°N; 76.6456° E)	Mr. Padmanbhan	Nov - Dec, 2013 Jan, 2014
Wayanad	Kalpetta (11.6198°N; 76.0843° E)	Mr. Baby	August, 2013
	Meenangady (11.6596°N; 76.1726° E)	Col. Madhavan Nair	August, 2013
	Mananthavady (11.8014°N; 76.0044° E)	Mr. Digaul Thomas	Aug - Sept, 2013 Dec, 2013 Jan – Feb, 2014
Thiruvananthapuram	Neyyatinkara (8.4016°N; 77.0871° E)	Mr. Cycyl Chandran	June, 2015
	Vellanad (8.5642°N; 77.0593° E)	Mr. Manoj	Nov- Dec, 2014 Jan- Feb, 2015 Apr- June, 2015

Palakkad

Wayanad 1

Wayanad 2

Wayanad 3

Thiruvananthapuram 1

Thiruvananthapuram 2

Plate 1. Polyhouses surveyed in Palakkad, Wayanad and Thiruvananthapuram districts

Thrissur 1

Thrissur 2

Thrissur 3

Thrissur 4

Thrissur 5

Plate 2. Polyhouses surveyed in Thrissur district

identification. Other mite specimens were mounted in the dorsal view. The slide mounted specimens were observed under phase contrast microscope (Leica DM 500) with attached image analyzer to study morphological characters. Identification of species was established using standard taxonomic keys (Ehara, 1995; Gupta and Gupta, 1999 and Srinivasa *et al*., 2012).

3.1.1.1. Molecular Characterization of Tetranychid Mites

For precise identification and confirmation of species of *Tetranychus* collected during the study, molecular characterization was carried out during January to April, 2014.

3.1.1.1.1. Maintenance of Isoline of Tetranychus sp.

Spider mites collected on cucumber from polyhouses of Thrissur district were used for maintaining isoline culture. Mite infested leaves collected during the survey were observed in laboratory under stereo zoom microscope and gravid female mites from each sample were transferred separately on to cucumber leaf bits in Petri Plates lined with wet cotton pad to maintain isoline culture (Plate 3). The leaf bits were replaced with fresh ones once in two days.

3.1.1.1.2. Morphological Characterization

Adult male and female mites, representing single species from the monoculture were slide mounted separately using Hoyer's media. Male specimens were also mounted in the lateral position to ensure better orientation of the genital structures, to study the aedeagal character for species determination. The slides were then labeled with details of host, locality and date of collection. Morphological characters of the slide mounted specimens were studied under phase contract research microscope and identity of the species was established based on the available literature (Gupta and Gupta, 1999; Ehara, 1995 and Srinivasa *et al*., 2012).

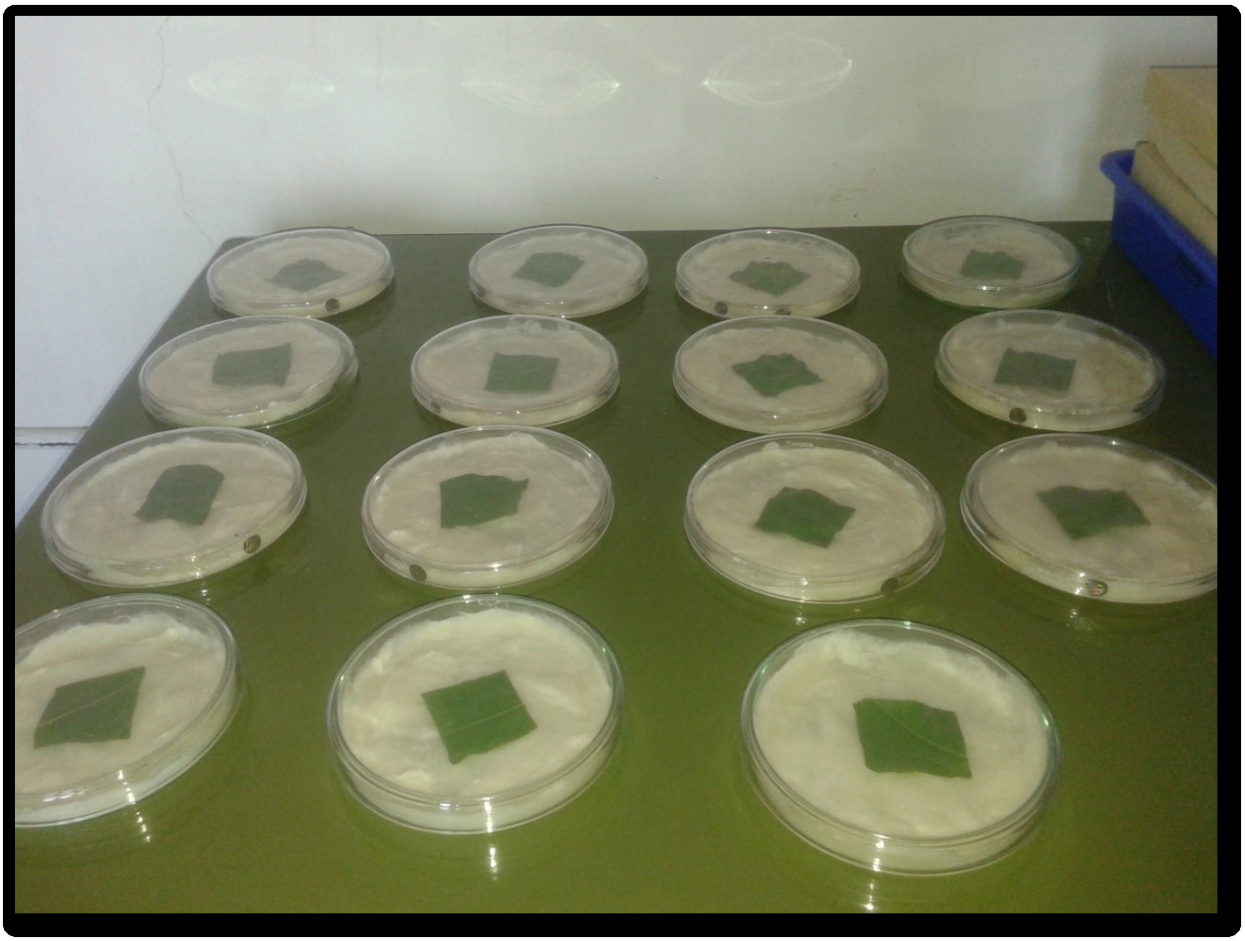


Plate 3. Isoline of *Tetranychus* sp.

3.1.1.1.3. Isolation and Characterization of Genomic DNA

Molecular characterization was carried out at the Centre for Plant Biotechnology and Molecular Biology, College of Horticulture, Vellanikkara. Ten adult females from the isoline maintained were used for DNA isolation. Genomic DNA was isolated using modified CTAB method. The polymerase chain reaction was used to amplify the second internal transcribed spacer (*ITS2*) of the ribosomal DNA and the cytochrome c oxidase subunit I (*COI*) locus of the mitochondrial genome. The amplified product was run on two per cent agarose gel. The quality and quantity of the eluted and purified DNA were confirmed using agarose gel electrophoresis and was further subjected to PCR with same primer sets to confirm that the band contains the amplified products from a single region of the genome. The PCR product, confirmed to yield only single band on electrophoresis, was further sent for sequencing with specific set of primers. The sequencing was carried out at Sci Genome Lab. Pvt. Ltd., Cochin. The DNA sequences obtained were blasted against GenBank (NCBI) to check the species identity in the database. Later, the sequences along with digital specimen photographs were submitted to BOLD.

3.1.2. Incidence and Crop Phenology Relationship of *Tetranychus urticae* on Cucumber

Fixed plot surveys were conducted at fortnightly intervals in five selected polyhouses in Thrissur district during July 2013 to May 2014, to record the incidence of *T. urticae* and document the relationship between crop growth stages and mite population in cucumber. To estimate the mite population at different growth stages, mite infested leaves (3 leaves/ plant) representing top, middle and bottom canopy were collected separately in polythene bags from ten randomly selected cucumber plants. In the laboratory, the leaves were observed under stereo binocular microscope and mite counts were recorded from three windows of 1 cm^2 area from each leaf. The average number of mites per cm^2 leaf area was estimated for different periods. The

data was subjected to ANOVA and the preference of spider mites to the crop stage was analyzed.

3.1.3. Identification of Natural Enemies of Phytophagous Mites

Predatory mites associated with the phytophagous mites on cucumber leaves, collected during the survey were slide mounted on Hoyer's media. Identification of slide mounted specimens was made with the support of appropriate literature (Gupta, 2003). Insect predators associated with the mites on cucumber were preserved in 70 per cent alcohol, labeled and sent to National Bureau of Agricultural Insect Resources, Bangaluru (NBAIR) as well as Biosystematics laboratory, Department of Agricultural Entomology, University of Agricultural Sciences, Bangaluru, for identification. During the study, mycosis of spider mites on cucumber was observed in a polyhouse. Isolation of the fungus from moribund mites was carried out in Sabouraud Dextrose Agar with the addition of two per cent Yeast extract medium (SDAY). The purified culture and mycosed mite specimens were sent for identification to National Center for Fungal Taxonomy (NCFT), New Delhi.

3.2. BIOLOGY OF *Tetranychus urticae* ON CUCUMBER

The study on the biology of two spotted spider mite, *T. urticae* on cucumber variety Hilton was conducted in the Acarology laboratory, College of Horticulture, Vellanikkara during June- July, 2013 at 27.05 ± 0.68ºC and 90.12 ±6.88 per cent relative humidity.

3.2.1. Mass Culturing of *Tetranychus urticae*

Tetranychus urticae was mass multiplied in the laboratory on cucumber leaves placed on plastic trays (40 x 28 cm^2) lined with well moistened synthetic absorbent sponge and a layer of blotting paper. Leaves were placed upside down on

the wet blotting paper and gravid females were released. Leaves were replaced with fresh ones once every two days.

3.2.2. Life History of *Tetranychus urticae* on Cucumber

The development and life history of *T. urticae* was studied following the leaf disc method (Krishna, 2013). Leaf discs of 4 cm^2 area were cut out from cucumber leaves and placed on wet cotton bed in Petri plates of 150 mm diameter. Three leaf bits were placed in a single Petri plate and 15 such Petri plates were maintained for the study (Plate 4). Gravid females were collected from mass culture and transferred to individual leaf discs at the rate of one gravid female per disc for oviposition. The gravid females and the excess eggs were removed from the leaf bit, 24 h later. Leaves were changed every two days to avoid poor nutrition for the mite.

3.2.2.1. Morphology and Developmental Duration of Immature Stages of Tetranychus urticae

The development of immature stages and morphology of different life stages of *T. urticae* were observed with the help of a stereo binocular microscope at two h interval until they reached maturity. The developmental duration of each stage namely, 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 ± Standard Deviation (SD).

3.2.2.2. 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 h later. The mated female was maintained till death. Leaf discs were changed at every three days interval. Ten

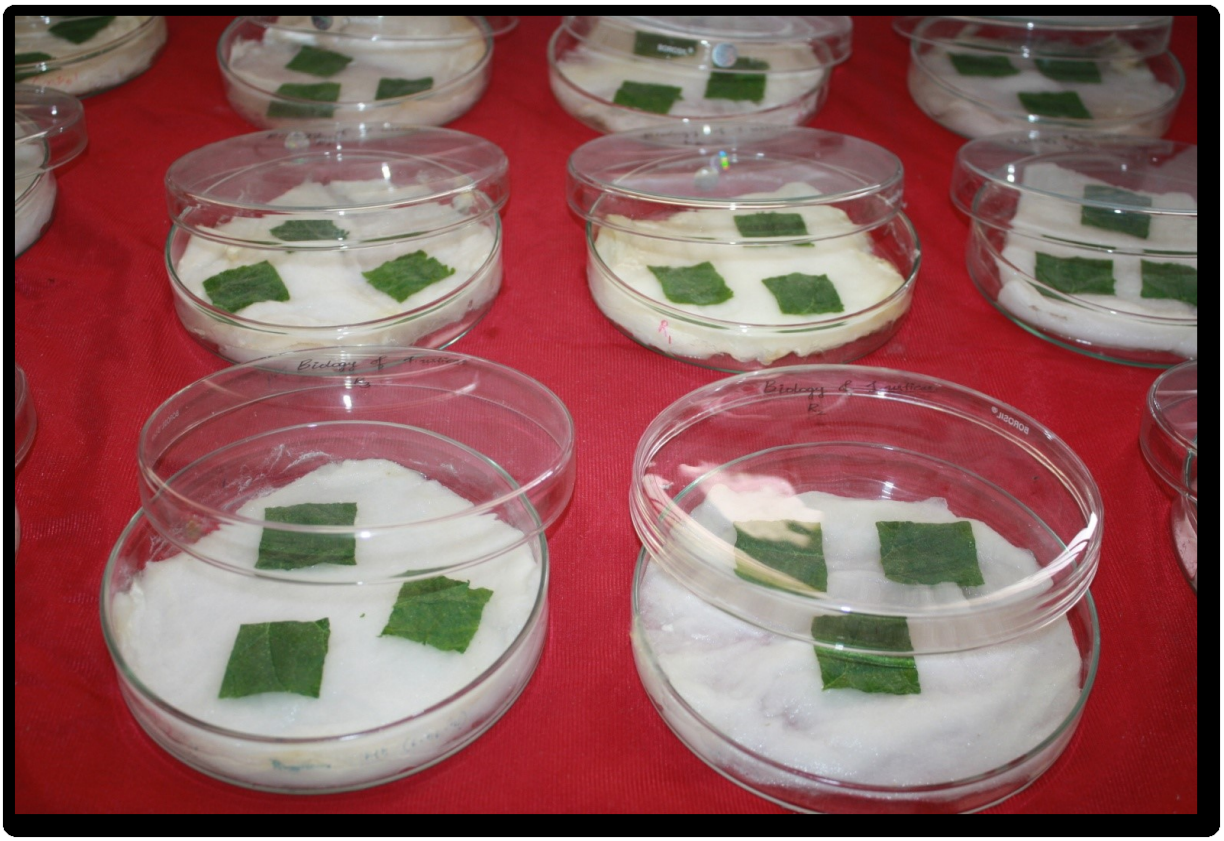


Plate 4. Leaf disc method for studying the biology of *Teranychus urticae*

replications each were maintained for males, mated females and unmated females to work out the longevity, expressed as mean days ± Standard Deviation (SD).

3.2.2.3. Reproductive Biology of Tetranychus urticae

To determine the duration of sexual development of mated female, one female teliochrysalis 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 h after the emergence of the female. The reproductive biology of unmated female was also studied using teliochrysalis that moulted to female but not allowed to mate. Ten replications each were maintained for mated and unmated females. Observations on pre- oviposition, oviposition and post- oviposition periods were recorded. The number of eggs laid by the mated as well as unmated females was recorded by replacing the leaf disc carrying eggs with fresh discs at two days interval till death of the female. The values were expressed as mean days ± Standard Deviation (SD). Fecundity was expressed as mean number of eggs per female ± Standard Deviation (SD).

3.2.2.4. Sex Ratio and Viability of Eggs

Sex ratio and viability of eggs were studied following the method described by Krishna (2013). 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.

3.2.2.5. Morphometry of Developmental Stages of Tetranychus urticae

Ten individuals each of different stages from egg to adult were randomly selected and morphometric parameters were recorded. The diameter of egg as well as maximum body length and width of other developmental stages were recorded in

micrometers (µm) using stereo zoom binocular microscope with HD camera attachment equipped with image analyzer software and expressed as mean values in mm ± Standard Deviation (SD).

3.3. BIOLOGY OF *Neoseiulus longispinosus* ON *Tetranychus urticae*

The studies on the biology of *N. longispinosus* on *T. urticae* were carried out during December- January, 2014 at 28.05 ± 0.90 ºC and 79.68 ± 8.11 per cent relative humidity, using cucumber variety Hilton

3.3.1. Field Collection and Identification of *Neoseiulus longispinosus*

The nucleus culture of *N. longispinosus* was maintained in the laboratory from the field collected native population. For this, gravid females collected from the vegetable seed production plot of Department of Olericulture, College of Horticulture were released separately on mulberry leaves infested with *T. urticae*. After five to six days, one male and female were selected at random from each leaf bit and mounted on glass slide. The slide mounted specimens were studied for morphological characters under stereo zoom microscope and species confirmed using standard taxonomic key (Gupta, 2003). The characters such as the shape of spermatheca and spermadactyl were considered for the same. This nucleus culture was used to mass culture *N. longispinosus* on *T. urticae* in the laboratory for the subsequent studies.

3.3.2. Laboratory Culture of *Neoseiulus longispinosus*

Neoseiulus longispinosus from the nucleus culture was multiplied in the laboratory on mulberry leaves infested with *T. urticae*. The mulberry leaves were maintained on plastic trays (40 x 28 cm^2) lined with well moistened synthetic absorbent sponge and a layer of absorbent cotton (Plate 5). Leaves were placed upside down on the moist cotton and gravid females of *T. urticae* were released. One week after the release of *T. urticae,* five gravid females of *N. longispinosus* were released on the leaf. Prey mites were replenished once every two days and the

mulberry leaves were replaced once in a week. For replacing mulberry leaf, old leaf was placed above the new leaf so that the prey and predatory mite got transferred to the new leaf naturally.

3.3.3. Mass Culturing of *Neoseiulus longispinosus*

Mass culture of *N. longispinosus* was maintained on cucumber plants (Variety Hilton) infested with *T. urticae* inside the polyhouse of AINPAA (Plate 6). The culture was initiated by releasing the gravid females from the laboratory culture.

3.3.4. Life History of *Neoseiulus longispinosus* on *Tetranychus urticae* on Cucumber

Developmental biology of *N. longispinosus* was studied on cucumber leaves infested with *T. urticae*. The experimental arena consisted of a plastic box (21.5 x 13.5 cm^2) having six cells of 7 x 7cm^2 each (Plate 7). Moist foam lined with cotton pad was placed in each cell and the cells were kept moist by adding water as and when required. Cucumber leaf bits of size 4 x 4 cm size were placed above the moist cotton pad in each cell (Plate 8). Twenty gravid females of *T. urticae* were transferred on to each leaf bit and were allowed to lay eggs. After 24 h, *T. urticae* females were removed and a single gravid *N. longispinosus* female was released on each leaf bit. The female was removed 24 h after egg laying. A single egg of *N. longispinosus* was maintained on each leaf bit for the study.

3.3.4.1. Morphology and Developmental Duration of Immature Stages of Neoseiulus longispinosus

The development of immature stages and morphology of different life stages of *N. longispinosus* was observed under stereo binocular microscope at two h interval until adult emergence. Leaf bits were changed and prey mites were replenished at two days interval. The developmental duration of each life stage, namely egg, larva and nymphal stages were recorded until adult emergence. On emergence the adult mites

Plate 5. Laboratory culture of *Neoseiulus longispinosus*

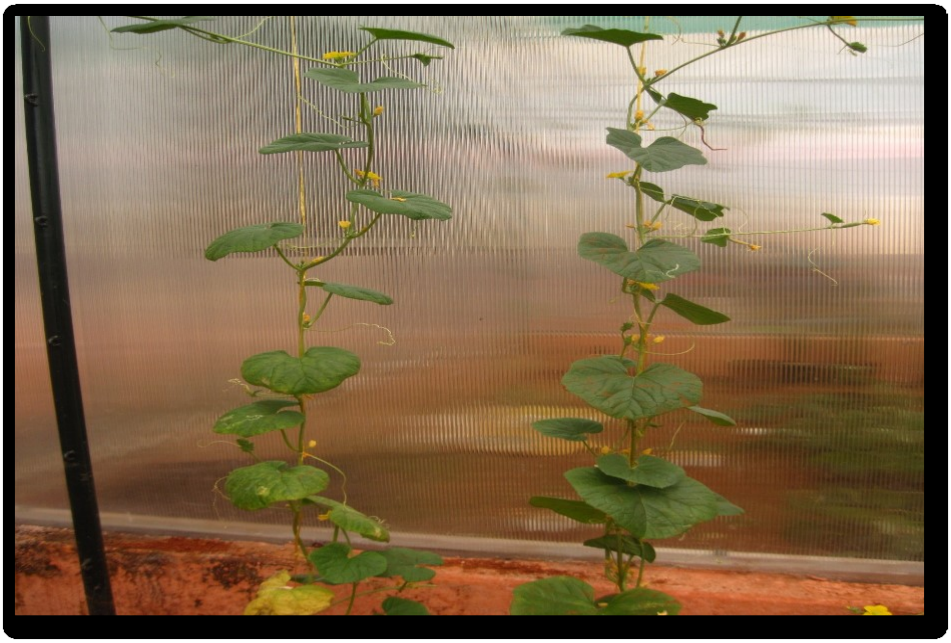


Plate 6. Mass culturing of *Neoseiulus longispinosus* on cucumber plants in polyhouse

Plate 7. Spice boxes used to study the biology of *Neoseiulus longispinosus*

Plate 8. Experimental set up for studying the biology of *Neoseiulus longispinosus*

were sexed out to work out the developmental duration of different stages separately for males and females. The values of developmental duration were expressed as mean days ± Standard Deviation (SD).

3.3.4.2. Adult Longevity of Neoseiulus longispinosus

Newly emerged males and females were maintained on separate leaf discs where they were retained until death to determine their longevity. Leaf discs were changed at every three days interval. Ten replications each were maintained for males and females to work out the longevity and expressed in mean days ± Standard Deviation (SD).

3.3.4.3. Reproductive Biology of Neoseiulus longispinosus

The experimental arena to study the reproductive biology of *N. longispinosus* was prepared as explained in 3.3.4. One female deutonymph of *N. longispinosus* was transferred to the leaf bit followed by the release of two adult males and allowed to mate after the final moult. The males were removed 24 h after the emergence of females. To study the reproductive biology of unmated female, single female deutonymph of *N. longispinosus* maintained separately and was not allowed to mate. Twelve replications were maintained for the study. Observations on pre- oviposition, oviposition and post oviposition were recorded. The number of eggs laid by the female was recorded by replacing the leaf discs carrying eggs with fresh discs till the death of the female. The values of pre- oviposition, oviposition and post oviposition periods were expressed as mean days ± Standard Deviation (SD). Fecundity was expressed as mean number of eggs per female ± Standard Deviation (SD).

3.3.4.4. Sex Ratio and Viability of Eggs

Eggs laid by a single female were reared and the viability was determined by counting the number of eggs that hatched out to larvae. From this, the per cent

viability of eggs was worked out. The emerging mites were sexed out after reaching adulthood to determine the sex ratio. Six replications were maintained for the study.

3.3.4.5. Morphometry of Developmental Stages of Neoseiulus longispinosus

Ten individuals each of different stages from egg to adult were randomly selected and morphometric parameters recorded. The maximum length and width of the egg and other developmental stages were recorded in micrometers (µm) using stereo zoom microscope with HD camera attachment equipped with image analyzer and expressed as mean ± Standard Deviation (SD).

3.4. EFFICACY AND PREY STAGE PREFERENCE OF *Neoseiulus longispinosus* ON *Tetranychus urticae*

Experiments were laid out in the laboratory to estimate the predatory potential, time needed to control the prey population and also to identify the preferred stage of the prey at 28.27 ± 0.25 º C and 80.25 ± 2.87 per cent relative humidity. The method adopted is explained below.

3.4.1. Predatory Potential of *Neoseiulus longispinosus* on *Tetranychus urticae*

Predatory potential is the number of prey consumed by a single predator in 24 h. Experiment on the predatory potential were carried out in plastic boxes having six cells of 7x7 cm^2 area, each cell being one replication. The cells were lined with absorbent sponge above which moist cotton pad was placed. Cucumber leaves of 4 x 4 cm^2 was kept on the moist cotton pad and the leaf bit was given cotton lining all around to prevent the escape of predators (Plate 9). The experiment was conducted at seven prey densities (10, 15, 20, 25, 30, 40 and 50) each of two prey stages namely, egg and active stages. Experiments were laid out separately for nymph and gravid female of *N. longispinosus,* with six replications for each treatment. To maintain the required prey density of egg, ten gravid females of *T. urticae* were released on to each leaf bit and were allowed to lay eggs. The females as well as the excess eggs

Plate 9. Experimental arena to study the predatory potential of *Neoseiulus longispinosus*

were removed 24 h later, after retaining the required number of eggs for each prey density. This was followed by the release of a single predator using a moist camel hair brush. To maintain the required prey density of active stages of the prey, mites were carefully transferred onto leaf discs with a zero size camel hair brush followed by a single predator. The predator was starved for three h prior to the experiment by confining them on a leaf bit without prey. After 24 h, the number of prey consumed was counted from each replication and recorded. The data was subjected to ANOVA and the means were compared using CD.

3.4.2. Estimation of Time Needed to Eliminate the Prey Population

The experimental arena consisted of excised leaf of cucumber kept on moist cotton pad placed in a Petri plate of diameter 150 mm. The petiole of the cucumber leaf was plugged with a thick cotton wad which was periodically moistened to keep the leaf turgid so that the leaf remained fresh for 10- 12 days (Plate 10). The experiment was conducted using gravid females of *T. urticae* at five different densities of 10, 15, 20, 25 and 30. A single gravid female of *N. longispinosus* was released on each leaf followed by the release of prey mites. The gravid female predator was starved for three h prior to the experiment by confining them on a leaf bit without prey. Cucumber leaf with gravid females of *T. urticae* at corresponding densities without predator served as control. All the life stages of the prey and predator in each treatment were counted daily until the number of prey becomes zero. Treatments were replicated four times. The Growth Rate (GR) of the prey and predator was worked out on the day when prey population reached zero. The growth rate was calculated as

GR = (Ending value/ Beginning value) ^ (1/n) -1*100

Where, n is the day at which prey population reaches zero.

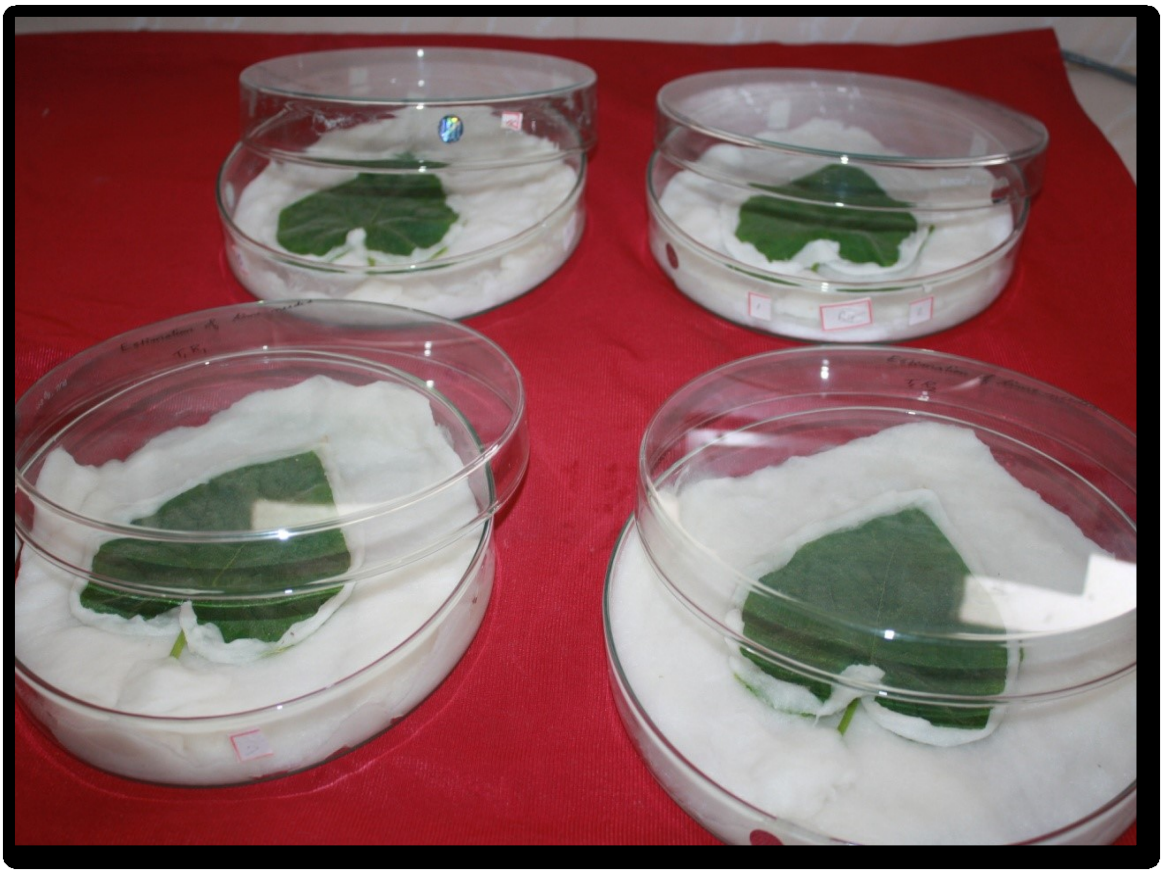


Plate 10. Experimental set up to study the time needed to control the prey population

3.4.3. Prey Stage Preference of *Neoseiulus longispinosus*

The experimental arena to study prey stage preference was set up as in experiment 3.4.1. Prey stage preference of *N. longispinosus* to different stages of *T. urticae* (egg, larva and nymph) was studied separately for nymph and gravid female. The study was conducted at three prey densities of 15, 30 and 45, each density comprising of equal number of different stages of the prey. Three gravid females of *T. urticae* were released on to each leaf bit and were allowed to lay eggs. After 24 h, the female as well as the excess eggs were removed after retaining the required number of eggs for each prey density. The required number of larvae and nymphs of *T. urticae* were then released on to the leaf bits carrying eggs using a zero size camel hair brush. Using a fine moist hair brush, a single predator (starved for 3 h) was transferred on to the experimental arena. Ten replications were maintained for the study. After 24 h, the predators were removed and the number of dead mites of each life stage was counted and the per cent consumption by each life stage was derived. The data was subjected to ANOVA and the means were compared using CD.

3.5. ESTIMATION OF PREDATOR: PREY RATIO OF *Neoseiulus longispinosus*

In order to recommend *N. longispinosus* for field application, the optimum number of predators required for controlling the known number of prey mites need to be standardized. To estimate the optimum predator: prey ratio, experiments were conducted in the laboratory as well as in polyhouse.

3.5.1. Estimation of Optimum Predator: Prey Ratio in Laboratory

The experiment was laid out in a Completely Randomized Design with eight treatments and three replications in laboratory during November, 2015 at 28.45 ± 0.63 º C and 69.79 ± 5.99 per cent relative humidity. Hundred mixed stages of *T. urticae* were released on cucumber leaves maintained on moist sponge in plastic trays (40 x 28 cm^2) (Plate 11). To prevent the movement of mites among the leaves in a

Plate 11. Experimental set up to study the optimum predator: prey ratio in laboratory

tray, a thin lining of wet cotton was provided all around the leaf margin. Using a moist hair brush, gravid females of *N. longispinosus* were released to at densities of 1, 2, 3, 4, 5, 10 and 20, separately. Cucumber leaf with hundred mixed stages of the prey without predator served as control. Number of eggs and active stages of both the prey and the predatorwas counted on 3rd, 7th and 10th day after release of the predator. The data were subjected to ANOVA and the means were compared using CD.

3.5.2. Estimation of Optimum Predator: Prey Ratio in Polyhouse

An experiment was conducted to evaluate the efficacy of *N. longispinosus* against *T. urticae* at four different predator: prey ratio (1: 20, 1: 25, 1: 33 and 1: 50) on cucumber (Variety Hilton) in the polyhouse of AINPAA during March to May, 2016. The crop was raised in the polyhouse as per the package of practices recommendations (KAU, 2014) at spacing of 60 x 30 cm in plots of 1.6 m x 1.3 m. The experiment was laid out in Completely Randomized Design with five treatments and four replications.

Hundred mixed stages of *T. urticae* were released on four leaves of twenty days old cucumber plants at the rate of 25 mites/leaf. This was followed by the release of gravid females of *N. longispinosus* at densities of five, four, three and two to obtain predator: prey ratios of 1: 20, 1: 25, 1: 33 and 1: 50. A control treatment was maintained without the release of the predator. Fifteen days after the first release, a second release of the predator was done at the same density.

Observations were recorded on mite population on 5, 10 and 15 days after the first and second release of the predator. Mite counts were recorded from three randomly selected mite infested leaves per plant. Number of mites/cm^2 leaf area was recorded *in situ* from five loci/leaf using a hand lens of 10 X magnification (Plate 12). Average number of mites/cm^2 leaf area was derived. The data were subjected to ANOVA.

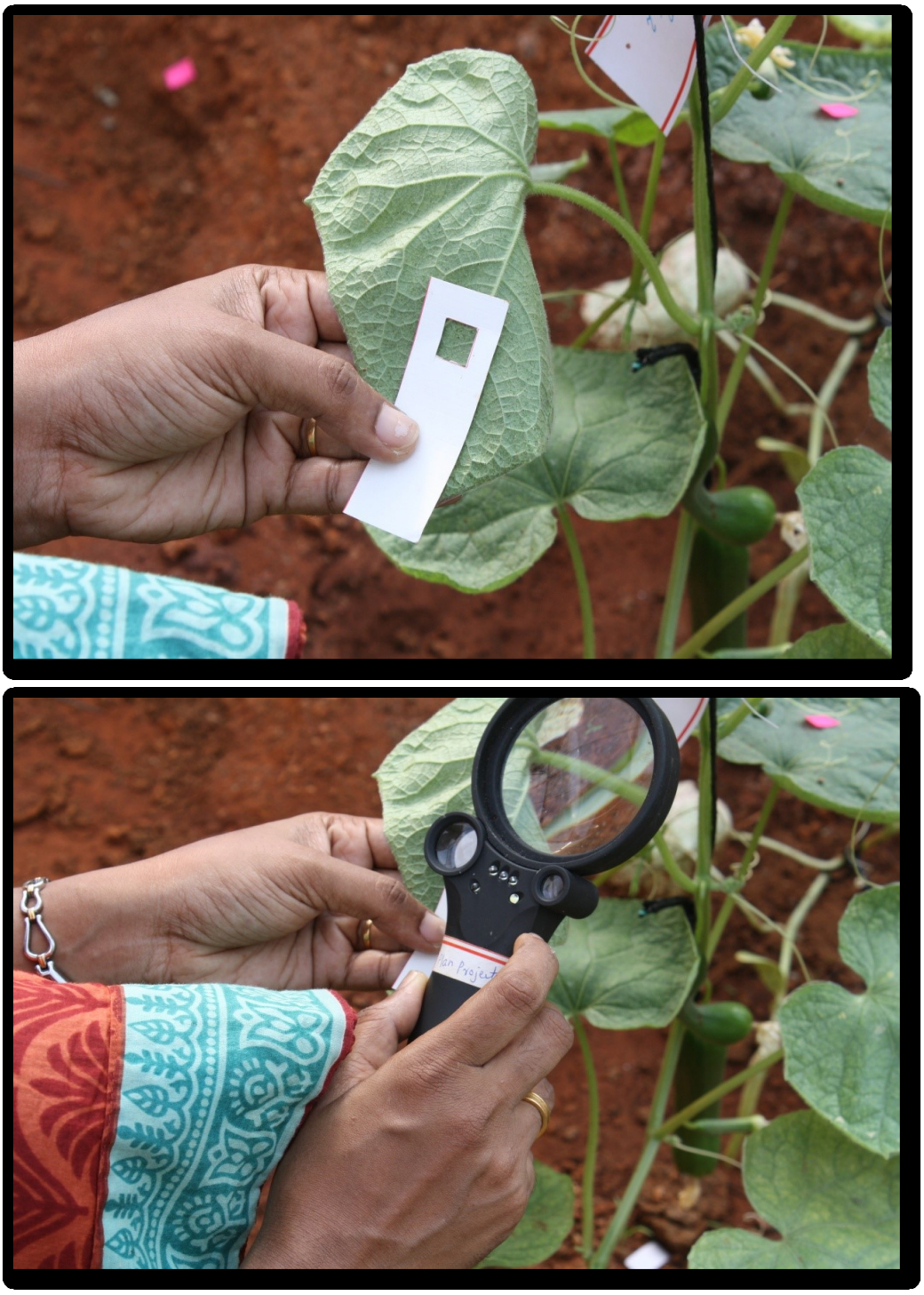


Plate 12. *In situ* counting of mites

Results

4. RESULTS

The results of the investigations carried out on the "Efficacy of *Neoseiulus longispinosus* (Evans) (Mesostigmata: Phytoseiidae) for the management of *Tetranychus urticae* Koch (Prostigmata: Tetranychidae) on cucumber under protected cultivation" are presented in this chapter.

4.1. INCIDENCE, CROP PHENOLOGY RELATIONSHIP AND NATURAL ENEMIES OF *Tetranychus urticae*

4.1.1. Diversity of Phytophagous Mites on Cucumber in Polyhouse

Roving surveys were undertaken to explore the mite fauna associated with cucumber in randomly selected fifteen polyhouses located in four districts of Kerala namely, Palakkad, Wayanad, Thiruvananthapuram and Thrissur. A total of five species of phytophagous mites belonging to two families were recorded on cucumber (Table 2). The mite families recorded were Tetranychidae represented by the genera *Tetranychus, Eutetranychus,* and Tarsonemidae, represented by *Polyphagotarsonemus.* Of the different species of spider mites on cucumber under protected cultivation, *Tetranychus truncatus* Ehara was found to be the predominant one. It was recorded from all the localities surveyed during the study. *Tetranychus urticae* was recorded only from Thrissur and Wayanad districts. *Tetranychus okinawanus* Ehara, an exotic pest was recorded on cucumber from one polyhouse located in Anthikkadu Block of Thrissur district. *Eutetranychus orientalis* (Klein) was observed in few numbers from two polyhouses of Thrissur district alone. The incidence of yellow mite, *Polyphagotarsonemus latus* (Banks) was also recorded from two polyhouses of Thrissur district.

Table 2. Phytophagous mites associated with cucumber in polyhouse

Sl. No.	Mite species	Family	District
1	*Tetranychus urticae* Koch	Tetranychidae	Thrissur, Wayanad
2	*Tetranychus truncatus* Ehara	Tetranychidae	Thrissur, Palakkad, Wayanad, Thiruvananthapuram
3	*Tetranychus okinawanus* Ehara	Tetranychidae	Thrissur
4	*Eutetranychus orientalis* (Klien)	Tetranychidae	Thrissur
5	*Polyphagotarsonemus latus* (Banks)	Tarsonemidae	Thrissur, Wayanad, Thiruvananthapuram

4.1.1.1. Molecular Characterization of Tetranychus spp.

Morphological characterization of mite specimens from the isoline culture revealed the presence of three species of *Tetranychus* namely *T. urticae* (Plate 13) *T. truncatus* (Plate 14) and *T. okinawanus* (Plate 15) on cucumber in polyhouses of Thrissur district. Molecular characterization confirmed the occurrence of the species, *T. truncatus* and *T. okinawanus* with GenBank accessions numbers, KT782744 (Plate 16) and KT782745 (Plate 17), respectively. Later the sequences along with the images of morphological characters were submitted to iBOL for barcoding. The process id received from BOLD are TOCRF001-15 for *T. truncatus* and TOIR001-15 for *T. okinawanus*.

4.1.2. Incidence and Crop phenology Relationship of *Tetranychus urticae* on Cucumber

The results of the study on incidence and crop phenology relationships showed that the population of spider mite was significantly higher during the late fruiting

Plate 13. *Tetranychus truncatus*

Plate 14. *Tetranychus urticae*

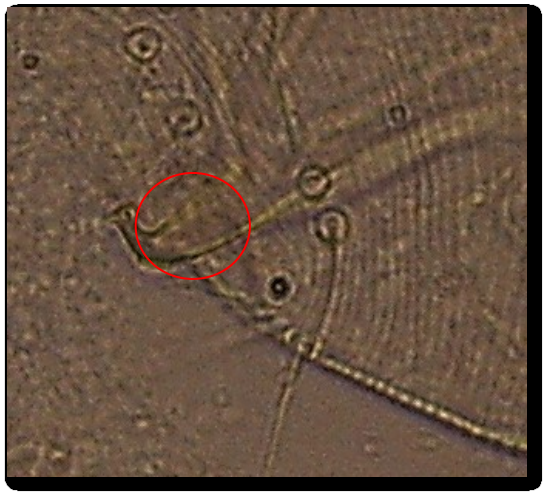
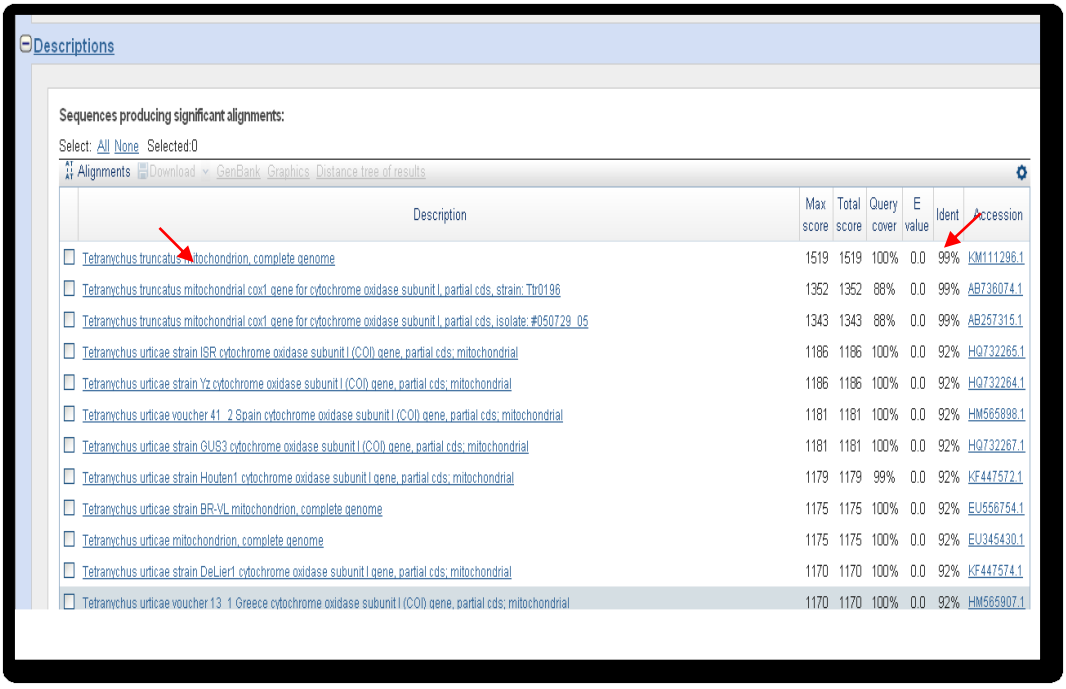


Plate 15. *Tetranychus okinawanus*

Plate 13 – 15. Aedeagus of *Tetranychus* spp.



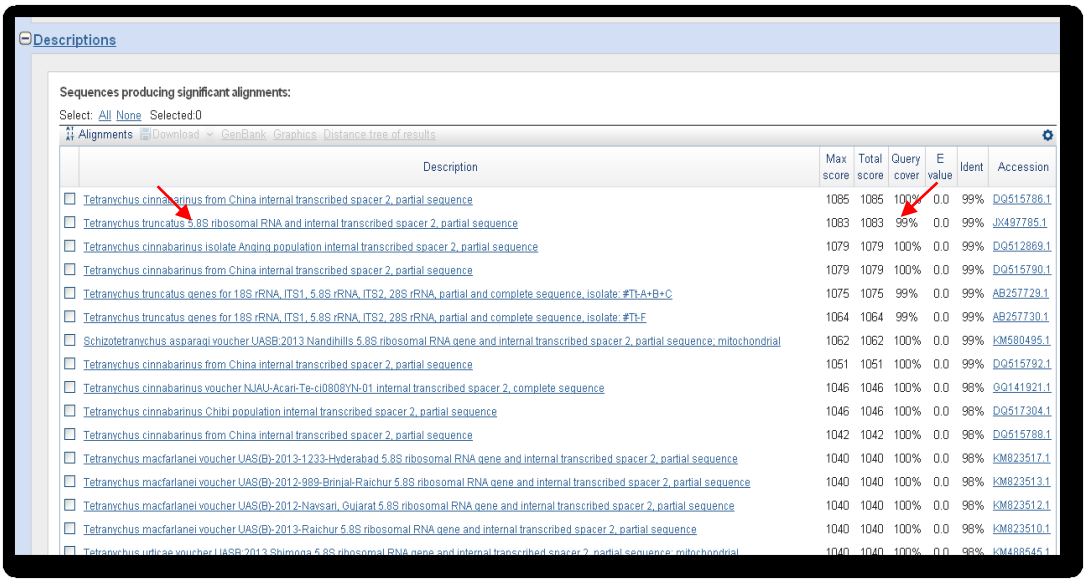
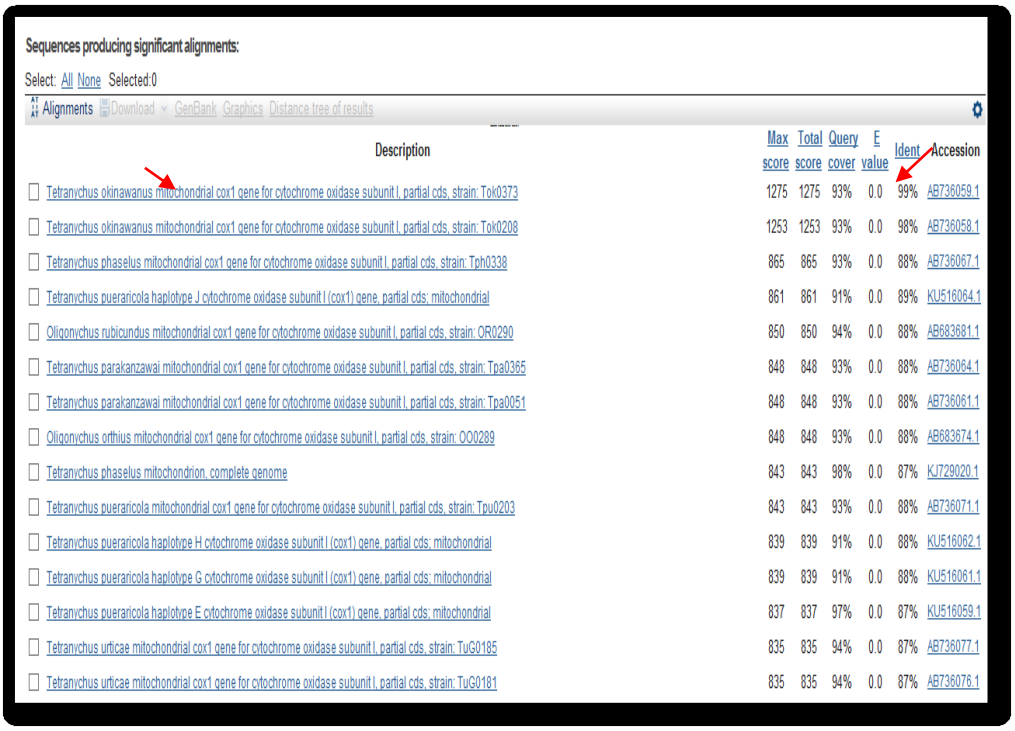


Plate 16. Blast result of *Tetranychus truncatus COI* and *ITS2*

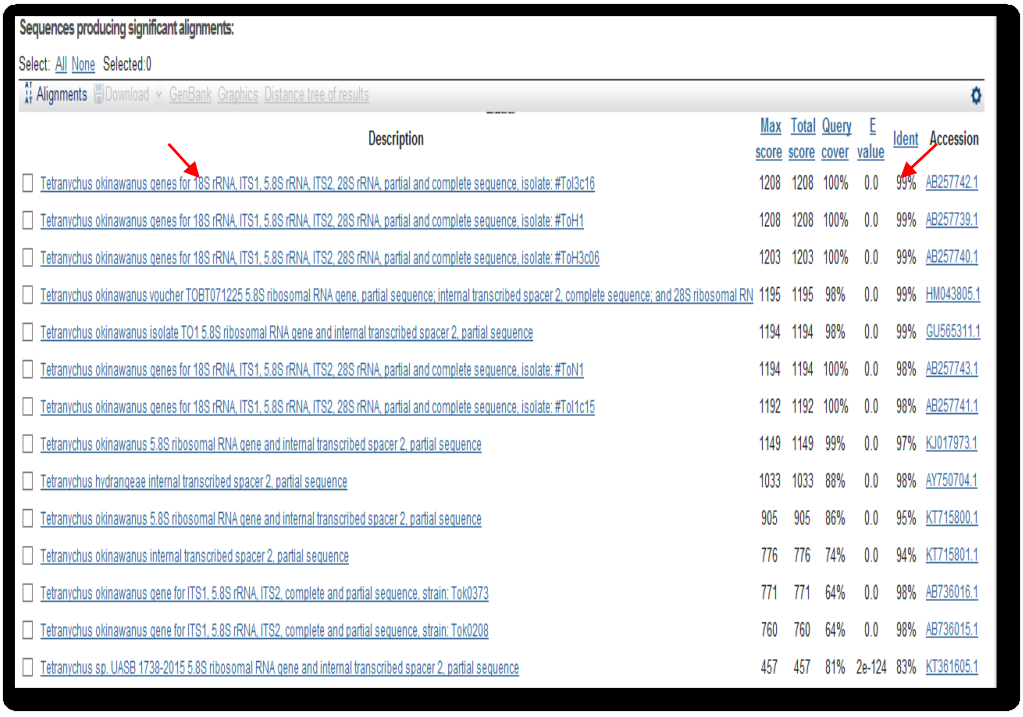


Sequences producing significant alignments:

Select: All None Selected:0

Alignments Download GenBank Graphics Distance tree of results

Description	Max score	Total score	Query cover	E value	Ident	Accession
Tetranychus okinawanus mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: Tok0373	1275	1275	93%	0.0	99%	AB736059.1
Tetranychus okinawanus mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: Tok0208	1253	1253	93%	0.0	98%	AB736058.1
Tetranychus phaselus mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: Tph0338	865	865	93%	0.0	88%	AB736067.1
Tetranychus pueraricola haplotype J cytochrome oxidase subunit I (cox1) gene, partial cds; mitochondrial	861	861	91%	0.0	89%	KU516064.1
Oligonychus rubicundus mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: OR0290	850	850	94%	0.0	88%	AB683681.1
Tetranychus parakanzawai mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: Tpa0365	848	848	93%	0.0	88%	AB736064.1
Tetranychus parakanzawai mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: Tpa0051	848	848	93%	0.0	88%	AB736061.1
Oligonychus orthius mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: OO0289	848	848	93%	0.0	88%	AB683674.1
Tetranychus phaselus mitochondrion, complete genome	843	843	98%	0.0	87%	KJ729020.1
Tetranychus pueraricola mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: Tpu0203	843	843	93%	0.0	88%	AB736071.1
Tetranychus pueraricola haplotype H cytochrome oxidase subunit I (cox1) gene, partial cds; mitochondrial	839	839	91%	0.0	88%	KU516062.1
Tetranychus pueraricola haplotype G cytochrome oxidase subunit I (cox1) gene, partial cds; mitochondrial	839	839	91%	0.0	88%	KU516061.1
Tetranychus pueraricola haplotype E cytochrome oxidase subunit I (cox1) gene, partial cds; mitochondrial	837	837	97%	0.0	87%	KU516059.1
Tetranychus urticae mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: TuG0185	835	835	94%	0.0	87%	AB736077.1
Tetranychus urticae mitochondrial cox1 gene for cytochrome oxidase subunit I, partial cds, strain: TuG0181	835	835	94%	0.0	87%	AB736076.1



Sequences producing significant alignments:

Select: All None Selected:0

Alignments Download GenBank Graphics Distance tree of results

Description	Max score	Total score	Query cover	E value	Ident	Accession
Tetranychus okinawanus genes for 18S rRNA, ITS1, 5.8S rRNA, ITS2, 28S rRNA, partial and complete sequence, isolate: #ToI3c16	1208	1208	100%	0.0	99%	AB257742.1
Tetranychus okinawanus genes for 18S rRNA, ITS1, 5.8S rRNA, ITS2, 28S rRNA, partial and complete sequence, isolate: #ToH1	1208	1208	100%	0.0	99%	AB257739.1
Tetranychus okinawanus genes for 18S rRNA, ITS1, 5.8S rRNA, ITS2, 28S rRNA, partial and complete sequence, isolate: #ToH3c06	1203	1203	100%	0.0	99%	AB257740.1
Tetranychus okinawanus voucher TOBT071225 5.8S ribosomal RNA gene, partial sequence; internal transcribed spacer 2, complete sequence; and 28S ribosomal RN	1195	1195	98%	0.0	99%	HM043805.1
Tetranychus okinawanus isolate TO1 5.8S ribosomal RNA gene and internal transcribed spacer 2, partial sequence	1194	1194	98%	0.0	99%	GU565311.1
Tetranychus okinawanus genes for 18S rRNA, ITS1, 5.8S rRNA, ITS2, 28S rRNA, partial and complete sequence, isolate: #ToN1	1194	1194	100%	0.0	98%	AB257743.1
Tetranychus okinawanus genes for 18S rRNA, ITS1, 5.8S rRNA, ITS2, 28S rRNA, partial and complete sequence, isolate: #ToI1c15	1192	1192	100%	0.0	98%	AB257741.1
Tetranychus okinawanus 5.8S ribosomal RNA gene and internal transcribed spacer 2, partial sequence	1149	1149	99%	0.0	97%	KJ017973.1
Tetranychus hydrangeae internal transcribed spacer 2, partial sequence	1033	1033	88%	0.0	98%	AY750704.1
Tetranychus okinawanus 5.8S ribosomal RNA gene and internal transcribed spacer 2, partial sequence	905	905	86%	0.0	95%	KT715800.1
Tetranychus okinawanus internal transcribed spacer 2, partial sequence	776	776	74%	0.0	94%	KT715801.1
Tetranychus okinawanus gene for ITS1, 5.8S rRNA, ITS2, complete and partial sequence, strain: Tok0373	771	771	64%	0.0	98%	AB736016.1
Tetranychus okinawanus gene for ITS1, 5.8S rRNA, ITS2, complete and partial sequence, strain: Tok0208	760	760	64%	0.0	98%	AB736015.1
Tetranychus sp. UASB 1738-2015 5.8S ribosomal RNA gene and internal transcribed spacer 2, partial sequence	457	457	81%	2e-124	83%	KT361605.1

Plate 17. Blast result of *Tetranychus okinawanus COI* and *ITS2*

stage of the crop (61-75 days after sowing), with an average of 21.12 mites in 1 cm^2 leaf area (Table 3). At early vegetative stage 13.95 mites were recorded per cm^2. The mite population during late vegetative stage (8.99 mites/ cm^2) and flowering stage (8.74 mites/ cm^2 were on par with the population at early vegetative stage. The lowest mite population of 3.82 mites per cm^2 was recorded during the early fruiting stage (46- 60 days), the same being significantly lower than mite population at all other stages.

4.1.3. Natural Enemies of Phytophagous Mites on Cucumber

A total of ten species of predators were found to be associated with spider mites during the survey of which, four were insect predators and six were mite predators (Table 4). The insect predators recorded were *Oligota* sp. (Coleoptera: Staphylinidae) (Plate 18a), *Stethorus pauperculus* (Weise) (Coleoptera: Coccinellidae) (Plate 18b), *Scolothrips* sp. (Thysanoptera: Thripidae) (Plate 18c) and an unidentified species of Cecidomyiidae (Plate 18 d). Predatory mites recorded included two species of Phytoseiidae as well as one species each of Stigmaeidae, Tydeidae, Cunaxidae and Cheyletidae. The predatory mite fauna associated with phytophagous mites on cucumber were *Agistemus garrulus* Chaudhari (Plate 19a), *Cheyletus* sp. (Plate 19b), *Cunaxa* sp. (Plate 19c), *Amblyseius paraaerialis* (Muma) (Plate 19d), *Neoseiulus longispinosus* (Evans) (Plate 19e) and *Tydeus gossabaensis* Gupta (Plate 19f). The phytoseiid predator, *N. longispinosus* was the predominant mite predator and was recorded from all the polyhouses surveyed during the study period. During the survey, mycosis of *T. urticae* was observed on cucumber grown in one polyhouse of Thrissur district during June, 2014. The fungus was isolated and identified as *Acremonium strictum* W. Gams (Plate 20). This is a new report on occurrence of natural epizootic of *A. strictum* on *T. urticae*.

Table 3. Population of mites on cucumber in polyhouse at different growth stages

Growth stages	***Mite population (Number of egg and active stages/cm^2 leaf area)**					
	Anthikkad	**Chavakkad 1**	**Chavakkad 2**	**Thanniyam**	**Vellanikkara**	**Mean**
Early vegetative (1- 15 days)	19.25 (4.44)	8.75 (3.04)	10.50 (3.31)	11.25 (3.42)	20.00 (4.52)	13.95^{b} (3.80)
Late vegetative (16- 30 days)	18.40 (4.34)	6.20 (2.58)	2.40 (1.70)	5.75 (2.50)	12.20 (3.56)	8.99^{bc} (3.08)
Flowering (31- 45 days)	6.00 (2.54)	4.33 (2.19)	5.50 (2.45)	11.50 (3.46)	16.40 (4.11)	8.74^{bc} (3.03)
Early fruiting (46- 60 days)	10.50 (3.31)	8.60 (3.01)	0.00 (0.70)	0.00 (0.70)	0.00 (0.70)	3.82^{c} (2.07)
Late fruiting (61- 75 days)	25.64 (5.11)	20.50 (4.58)	25.00 (5.04)	15.25 (3.96)	19.25 (4.44)	21.12^{a} (4.64)
CD						6.96

*Mean of 90 observations ** Figures in parenthesis are square root transformed values

Table 4. Natural enemies associated with spider mites on cucumber in polyhouses

Sl. No.	Species	Family	Order
1	*Stethorus pauperculus* (Weise)	Coccinellidae	Coleoptera
2	*Oligota sp.*	Staphylinidae	Coleoptera
3	*Scolothrips sp.*	Thripidae	Thysanoptera
4	Unidentified species	Cecidomyiidae	Diptera
5	*Neoseiulus longispinosus* (Evans)	Phytoseiidae	Mesostigmata
6	*Amblyseius paraaerialis* (Muma)	Phytoseiidae	Mesostigmata
7	*Agistemus garrulus* Chaudhari	Stigmaeidae	Prostigmata
8	*Tydeus gossabaensis* Gupta	Tydeidae	Prostigmata
9	*Cunaxa* sp.	Cunaxidae	Prostigmata
10	*Cheyletus* sp.	Cheyletidae	Prostigmata

4.2. BIOLOGY OF *Tetranychus urticae* ON CUCUMBER

The studies on the biology of *T. urticae* were conducted at 27.05 ± 0.68ºC and 90.12 ± 6.88 per cent relative humidity in the Acarology laboratory, Department of Agricultural Entomology, College of Horticulture, Vellanikkara during June- July, 2013.

a. *Oligota* sp. - Adult

Oligota sp. - Grub

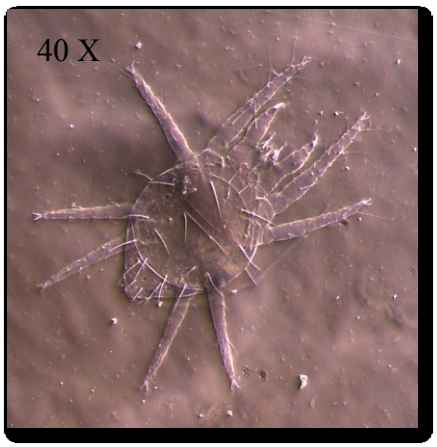
b. *Stethorus pauperculus* Weise- Adult

Stethorus pauperculus Weise- Grub

c. *Scolothrips* sp. - nymph

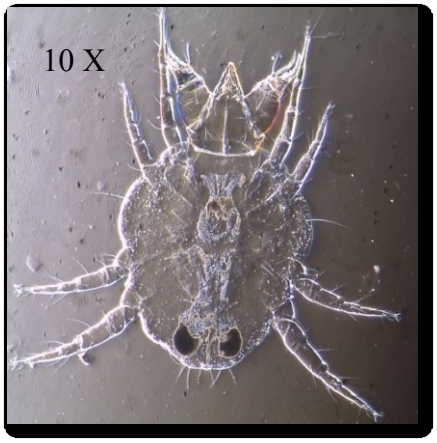
d. Cecidomyiidae – maggot

Plate 18 a- d. Insect predators of spider mites on cucumber

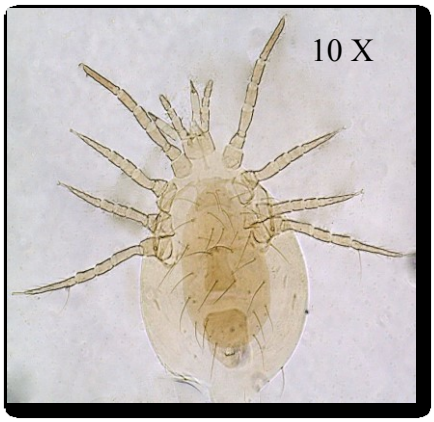


a. *Agistemus garrulous* Chaudhari

d. *Amblyseius paraaelialis* (Muma)

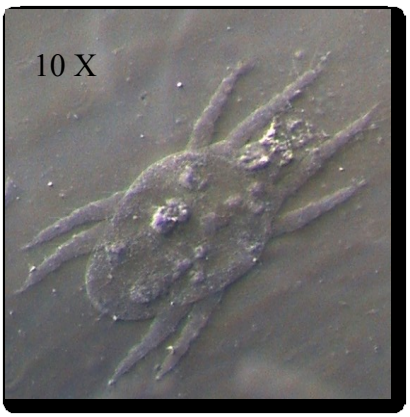


a. *Cheyletus* sp. (Evans)



e. *Neoseiulus longispinosus*

b. *Cunaxa* sp.



f. *Tydeus gossabaensis* Gupta

Plate 19 a- f. Predatory mite fauna associated with spider mites on cucumber

Plate 20. ***Acremonium strictum*** **infected mycosed mite**

4.2.1. Life History of *Tetranychus urticae* on Cucumber

The life cycle of *T. urticae* consisted of five different stages such as egg, larva, protonymph, deutonymph and the adult (Plate 21 a- f). Three short quiescent intervals called nymphochrysalis, deutochrysalis and teliochrysalis were present in between each life stage until deutonymph stage.

4.2.1.1. Morphology and Developmental Durations of Immature Stages of Tetranychus urticae

The morphological characters and the duration of development of various life stages of *T. urticae* recorded during the study is presented in Table 5.

4.2.1.1.1. Egg

Eggs were laid on the underside of the leaves, singly or in groups, on the web or on leaf, near the veins and midrib. The eggs were spherical in shape, translucent and clear when freshly laid but turned dark brown later on. Prior to hatching, the egg turned creamy white and two dark eye spots were very clear. This corresponds to the simple eyes of the larva. The mean incubation period for male was longer (2.85 days) compared to that of female (2.48 days).

4.2.1.1.2. Larva

The egg hatched out to larva. The larva possessed three pairs of legs. On hatching it was cream coloured, changing to pale green on feeding. The simple eyes on the dorso- lateral idiosoma were clearly distinguishable at this stage. The mean larval period recorded was 0.56 days for male and 0.70 days for female.

4.2.1.1.3. Nymphochrysalis

Nymphochrysalis is the quiescent stage that followed the larval stage. At this stage, larva stopped feeding and entered the resting stage with anterior pairs of legs

a. Egg

b. Larva

c. Protonymph

d. Deutonymph

e. Adult female

f. Adult male

*All magnifications in 35 X

Plate 21. Life stages of *Tetranychus urticae*

extended straight forward and kept close to each other and the postereor legs extended backwards and held close to the sides of opisthosoma. Average nymphochrysalis period was 0.80 days for male and 0.85 days for female.

4.2.1.1.4. *Protonymph*

Nymphochrysalis moulted to the first nymphal stage called protonymph. Protonymph was characterized by the presence of four pairs of legs. It was larger in size and darker in colour as compared to the larva. The mean protonymph period lasted for 0.57 days for male and 0.52 days for female.

Table 5. Duration of developmental stages of *Tetranychus urticae* on cucumber

Life stage	***Development period (Days ± SD)**	
	Male	**Female**
Egg	2.85±0.08	2.48±0.06
Larva	0.56±0.04	0.7±0.04
Nymphochrysalis	0.8±0.06	0.85±0.03
Protonymph	0.57±0.03	0.52±0.04
Deutochrysalis	0.64±0.04	0.78±0.03
Deutonymph	0.71±0.03	0.77±0.02
Teleiochrysalis	0.61±0.04	1.05±0.03
Total development period	6.75±0.11	7.15±0.11

*Mean of 20 observations

4.2.1.1.5. Deutochrysalis

The protonymph entered into quiescent stage, the deutochrysalis. It remained anchored to the leaf surface in a similar fashion as that of nymphochrysalis. This stage, on an average, lasted for 0.64 days in the case of male and 0.78 days in the case of female.

4.2.1.1.6. Deutonymph

Deutochrysalis moulted to the second nymphal stage, the deutonymph. Deutonymph was light reddish in colour. Female deutonymph was larger compared to male deutonymph which was elongate. The stage was found to be moving and feeding very actively. The mean deutonymphal period was 0.71 days for male and 0.77 days for female.

4.2.1.1.7. Teleiochrysalis

The deutonymph entered the third quiescent stage, the teleiochrysalis. This period on an average was 0.61 days for male and 1.05 days for female.

4.2.1.1.8. Adult

Teleiochrysalis moulted to adult mite. Sexual dimorphism was exhibited by the adults in size, shape and colour. The male was smaller with postereorly tapering body, whereas female was larger and plumpy with body rounded postereorly. The setae on idiosoma and legs of female were longer compared to those of male. The male is light red and female carmine red in colour. Females turned darker after mating. Both male and female had bright red eye spots on the dorso- lateral idiosoma.

4.2.1.1.9. Total development period

The total development period from egg to adult emergence was 6.75 days for male and 7.15 days for female.

4.2.1.2. Adult longevity

Adult male recorded a mean longevity of 8.95 days while mated and unmated female recorded longevity of 11.59 days and 13.04 days, respectively (Table 6).

4.2.1.3. Reproductive Biology of *Tetranychus urticae*

4.2.1.3.1. Pre- oviposition, oviposition and post- oviposition period

The pre- oviposition period, oviposition period and post- oviposition period, were observed to be of longer duration in unmated female compared to mated female. The mean pre-oviposition period in mated and unmated females lasted for 2.16 days and 1.78 days, respectively. Mean oviposition and post- oviposition periods lasted for 8.15 days and 1.67 days in case of mated females and 10.16 days and 1.86 days in case of unmated females as presented in Table 7.

4.2.1.3.2. Fecundity, Sex Ratio and Egg Viability of Tetranychus urticae

Mated female on an average laid 47.91 eggs whereas unmated female laid only 36.08 eggs. The progeny of mated female consisted of both males and females in the ratio 1:4.6, while unmated female produced only males. The viability of eggs of *T. urticae* was 92.55 per cent for mated female and 90. 23 per cent for unmated female (Table 8).

Table 6. Adult longevity of *Tetranychus urticae*

Sex	***Duration (Days ±SD)**
Male	8.95±0.17
Mated female	11.59±0.09
Unmated female	13.04±0.20

*Mean of 12 observations

Table 7. Pre- oviposition, oviposition and post- oviposition periods of *Tetranychus urticae*

Parameter	***Duration (Days ±SD)**	
	Mated female	Unmated female
Pre- oviposition	2.16±0.03	1.78±0.05
Oviposition	8.15±0.04	10.16±0.03
Post- oviposition	1.67±0.02	1.86±0.04

*Mean of 12 observations

Table 8. Fecundity, sex ratio and viability of *Tetranychus urticae*

	***Fecundity**	***Egg viability (%)**	***Male : Female ratio**
Unmated female	36.08±7.67	90.23±3.35	1:0
Mated female	47.91±2.81	92.55±1.48	1: 4.6

*Mean of 12 observations

4.2.2. Morphometry of Developmental Stages of *Tetranychus urticae*

The mite showed a gradual increase in size from egg to adult stage except that of teleiochrysalis which shrunk in size as compared to its deutonymph. As sex could be distinguished only from deutonymph stage, the measurements up to this stage were recorded in common. Eggs on an average measured 126.32 µm in diameter. The mean body length and body width of larva were 178.26 µm and 121.20 µm. Nymphochrysalis measured 218.11 µm in mean length and 129.56 µm in mean width. The length of the protonymph was 258.54 µm while the width on an average was recorded as 142.24 µm. The deutochrysalis measured 287.65 µm in length and 166.89 µm in width. Male deutonymph had a length of 356.31 µm and a width of 165.31 µm where as female deutonymph on an average measured 410.35 µm in length and 209.89 µm in width. Length and width of teleiochrysalis was 335.97 µm and 170.43 µm for male and 408.62 µm and 197.59 µm for female, respectively. The length and width of adult mite were 347.49 µm and 160.66 µm for male and 490.26 µm and 240.15 µm for female, respectively (Table 9).

Table 9. Morphometry of developmental stages of *Tetranychus urticae*

Life stage	***Length (µm) ± SD**		***Width (µm) ± SD**	
Egg	126.32 ± 1.60			
Larva	178.26 ± 1.94		121.20 ± 0.50	
Nymphochrysalis	218.11 ± 2.40		129.56 ± 1.60	
Protonymph	258.54 ±2.04		142.24 ± 2.20	
Deutochrysalis	287.65 ± 1.30		166.89 ± 2.83	
	Male		**Female**	
	***Length (µm)**	***Width (µm)**	***Length (µm)**	***Width (µm)**
Deutonymph	356.31 ± 2.03	165.31 ± 1.54	410.35 ± 5.30	209.89 ± 4.30
Teleochrysalis	335.97 ± 1.61	170.43 ± 2.16	408.62 ± 4.20	197.59 ± 2.69
Adult	347.49 ± 2.50	160.66 ± 3.25	490.26 ± 2.98	240.15 ± 3.18

*Mean of 10 observations

4.3. BIOLOGY OF *Neoseiulus longispinosus* ON *Tetranychus urticae* ON CUCUMBER

4.3.1. Life History of *Neoseiulus longispinosus* on *Tetranychus urticae* on Cucumber

The life cycle of *N. longispinosus* consisted of five different stages such as egg, larva, protonymph, deutonymph and the adult (Plate 22).

4.3.1.1. Morphology and Developmental Duration of Immature Stages of Neoseiulus longispinosus

The morphological characters and the duration of development of various life stages of *N. longispinosus* are presented in Table 10.

Table 10. Duration of developmental stages of *Neoseiulus longispinosus* on *Tetranychus urticae*

Stages/Period	***Male (Days ±SD)**	***Female (Days ±SD)**
Egg	1.46±0.02	1.57±0.13
Larva	0.60±0.04	0.65±0.02
Protonymph	0.86±0.03	0.98±0.04
Deutonymph	0.98±0.04	1.05±0.03
Total development period	3.91±0.05	4.27±0.13
Adult longevity	19.66±1.07	22.75±0.75

*Mean of 12 observations

a. Egg

b. Larva

b. Protonymph

d. Deutonymph

e. Adult female

f. Adult male

*All magnifications in 35 X

Plate 22. Life stages of *Neoseiulus longispinosus*

4.3.1.1.1. Egg

Neoseiulus longispinosus preferred to lay eggs on the underside of the leaf. Gravid female mite laid eggs singly or in groups, amidst *T. urticae* eggs. Egg is oval in shape and shiny white translucent when freshly laid. Later the colour turned dirty white or turbid, prior to hatching. The average incubation period was 1.46 days for male and 1.57 days for female.

4.3.1.1.2. Larva

Newly hatched larva is shiny white with three pairs of legs and is small in size. Larva is a non feeding stage but moved slowly around on the leaf. The mean larval period recorded was 0.60 days for male and 0.65 days for female.

4.3.1.1.3. Protonymph

The larva moulted into a protonymph which is a very active stage. Protonymph is characterized by four pairs of legs and was larger in size. It fed actively on *T. urticae*. The protonymph is dark greenish in colour initially, but later turned to pale orange. The mean protonymph period lasted for 0.86 days for male and 0.98 days for female.

4.3.1.1.4. Deutonymph

Protonymph moulted to the second nymphal stage, the deutonymph, a very actively moving and feeding stage. Deutonymph is bright orange in colour with brownish tint. The typical orange sclerotization pattern was visible at this stage. Female deutonymph is larger and wider than its male counterpart which was elongate. Sexual characters could be distinguished at this stage. The mean deutonymph period was 0.98 days for male and 1.05 days for female.

4.3.1.1.5. Adult

Deutonymph moulted to adult. The adult mite exhibited sexual dimorphism in colour, size and shape. Male was light brown in colour and smaller in size compared to the female. The shape of male was elongate and the hysterosoma was oblong, more or less like the base of a test tube. Female was dark brown in colour and bigger in size. The hysterosoma of female is broadly rounded. Older female was more plumby and darker in colour.

4.3.1.1.6. Total development period

The total mean development period from egg to adult emergence was 3.91 days for male and 4.27 days for female.

4.3.1.2. Adult longevity

Adult male recorded a mean longevity of 19.66 days while female lived on an average for 22.75 days.

4.3.1.3. Reproductive Biology of Neoseiulus longispinosus

4.3.1.3.1. Mating behaviour

Newly emerged male was found actively moving in search of females to mate. The male mated with female immediately after moulting. The female carried the male on its ventral side and the mating lasted for more than a minute (Plate 23). Male and female were observed to mate more than once in their life time. Parthenogenesis was not observed.

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

The mean pre-oviposition, oviposition and post- oviposition periods of mated female was 1.97, 19.91 and 0.99 days, respectively (Table 11). Unmated female did not lay eggs.

a. Mating by freshly emerged female

b. Mating by gravid female

Plate 23. Multiple mating in *Neoseiulus longispinosus*

4.3.1.3.3. Fecundity, sex ratio and egg viability of Neoseiulus longispinosus

Female, on an average, laid 31.33 eggs, of which, 89.83 per cent hatched out to larva. The progeny consisted of both males and females in the ratio 1: 3.31 (Table 11).

Table11. Biological parameters of *Neoseiulus longispinosus*

Parameter	***Duration (Days±SD)**
Fecundity	31.33±2.27
Viability (%)	89.30±2.47
Pre- oviposition period	1.97±0.03
Oviposition period	19.91±0.66
Post- oviposition period	0.99±0.02
Sex ratio (Male: female)	1: 3.31

*Mean of 12 observations

4.3.2. Morphometry of Developmental Stages of *Neoseiulus longispinosus*

The mite showed a gradual increase in size from larva to adult stage. Egg on an average measured 196.23 µm in length and 151.01 µm in width. The mean body length and width of larva were 212.60 µm and 182.30 µm, respectively. The body length of the protonymph measured 267.40 µm while the width was 202.06 µm. Deutonymph recorded a body length of 311.20 µm and width of 216.03 µm. The maximum length and width of adult mite were 426.86 µm and 227.58 µm for male and 441.30 µm and 260.86 µm for female, respectively (Table 12).

Table 12. Morphometry of developmental stages of *Neoseiulus longispinosus*

Life stage	***Length (µm) ± SD**	***Width (µm) ± SD**
Egg	196.23 ± 4.50	151.01 ± 2.01
Larva	212.60 ± 2.10	182.30 ± 1.36
Protonymph	267.40 ± 1.04	202.06 ± 1.16
Deutonymph	311.20 ± 3.60	216.03 ± 1.50
Adult male	426.86 ± 3.30	227.58 ± 1.80
Adult female	441.30 ± 2.70	260.86 ± 2.65

* Mean of 10 observations

4.4. EFFICACY AND PREY STAGE PREFERENCE OF *Neoseiulus longispinosus*

Experiments were conducted in the laboratory to estimate the predatory potential of *N. longispinosus*, time needed to control the prey population and also to identify the preferred stage of the prey, *T. urticae* at 28.27 ± 0.25 º C and 80.25 ± 2.87 per cent relative humidity.

4.4.1. Predatory Potential

The predatory potential of nymph and adult females of *N. longispinosus* were studied separately on eggs and active stages of *T. urticae*.

The predatory potential of deutonymph of *N. longispinosus* on eggs and active stages of *T. urticae* is presented in Table 13. The deutonymphs of *N. longispinosus* consumed the highest number of prey eggs at a prey density of 30 (7.67) This was followed by mean consumption of 7.00 and 6.83 eggs at prey densities of 40 and 25, respectively. All the above values were on par with each other and significantly superior to the consumption at other prey densities evaluated. Prey

consumption at the highest prey density of 50 mites was 6.67, which was at par with the corresponding value of 6.00 at 25 mites. The lowest consumption recorded was 2.33 eggs in 24 h, at a prey density of 10 which was on par with 2.83 eggs at a prey density of 15.

Table 13. Predatory potential of *Neoseiulus longispinosus* on *Tetranychus urticae* in 24 h at different prey densities

Prey density per predator	***Rate of predation (Number)**			
	Nymph of *N. longispinosus*		**Adult *N. longispinosus***	
	Egg	**Active stage**	**Egg**	**Active stage**
10	2.33^{d} (1.68)	1.33^{d} (1.35)	3.83^{e} (2.08)	1.83^{d} (1.52)
15	2.83^{d} (1.82)	1.33^{d} (1.35)	3.33^{e} (1.95)	2.00^{d} (1.58)
20	6.00^{c} (2.54)	2.33^{c} (1.68)	6.17^{d} (2.58)	3.67^{c} (2.04)
25	6.83^{abc} (2.70)	3.50^{b} (2.00)	7.17^{cd} (2.76)	5.00^{b} (2.34)
30	7.67^{a} (2.85)	3.83^{b} (2.08)	8.17^{c} (2.94)	4.50^{bc} (2.23)
40	7.00^{ab} (2.73)	5.67^{a} (2.48)	11.17^{a} (3.41)	7.83^{a} (2.88)
50	6.67^{bc} (2.67)	5.83^{a} (2.51)	9.83^{b} (3.21)	7.83^{a} (2.88)
CD	0.92	0.70	1.02	0.92

*Mean of six observations ** Figures in parenthesis are square root transformed values

The deutonymphs of *N. longispinosus* consumed the highest number of prey nymphs at a prey density of 50 (5.83). This was followed by mean consumption of 5.67 nymphs at prey densities of 40. These two values were on par with each other and significantly superior to the consumption at other prey densities evaluated. Prey consumption at prey density of 30 and 25 mites was 3.83 and 3.50 nymphs, which was at par with the each other. This was followed by the consumption at a prey density of 20 (2.33). The lowest consumption recorded was 1.33 nymphs in 24 h, at a prey density of 10 and 15.

The adult predator consumed significantly higher number of prey egg (11.17) at a prey density of 40. This was followed by the prey density of 50 (9.83 eggs). The predator adult consumed on an average 8.17 eggs at density of 30 which was on par with that consumed at a prey density of 25 (7.17 egg). At a prey density of 20, the predator adult consumed 6.17 eggs in 24 h. However this was significantly lower than that consumed at a prey density of 25. At a prey density of 10, it consumed on an average 3.83 eggs. Significantly lower consumption of prey eggs was recorded at the prey density of 15 (3.33).

The predator adult consumed significantly higher number of deutonymph (7.83) at prey densities of 50 and 40. At a prey density of 25, the predator adult consumed 5.00 deutonymphs in 24 h. However, the prey consumption at a prey density of 30 (4.50 deutonymph) was on par with that at a prey density of 25. This was followed by the prey density of 20 (3.67 deutonymph). The adult consumed on an average 2.00 and 1.83 deutonymph at prey densities of 15 and 10, respectively which were on par with each other.

4.4.2. Estimation of Time Needed to Eliminate the Prey

To estimate the time needed to devour the available prey population, a single gravid female of *N. longispinosus* was provided with gravid females of *T. urticae* at five different prey densities namely 10, 15, 20, 25 and 30. The population of both prey and predator was recorded daily. The results are presented in Tables 14, 15 and 16.

At an initial prey density of 10, the predatory mite took eight days to completely eliminate the available prey. At prey densities of 15, 20, 25 and 30, the available prey mite was completely devoured by the single predator in 11, 14, 16 and 18 days, respectively. When the prey mite population reached zero in the treatment, in the control, the population of prey mite at prey densities of 10, 15, 20, 25 and 30 were 121.75, 364.25, 536.50, 706.00 and 702.00. The result clearly indicated that the time needed to control the prey increased with increase in density of the prey (Table 14).

The reduced growth rate of prey mites in the arena with predator at different prey densities of 10, 15, 20, 25 and 30 recorded was 34.81, 14.18, 10.84, 11.14 and 11.51 per cent, respectively. The corresponding population growth rate of *T. urticae* in the control arena *i.e.* without predator was 37.83, 36.33, 28.11, 24.61 and 20.13 per cent, respectively at prey densities of 10, 15, 20, 25 and 30. The population growth rate of predator at different prey densities of 10, 15, 20, 25 and 30 were 21.90, 31.10, 23.47, 19.65 and 18.13, respectively (Table 16).

Table 14. Population of *Tetranychus urticae* after release of a single predator at different prey densities

Days	***Prey population (Number per leaf)**									
	10		**15**		**20**		**25**		**30**	
	W	**WP**	**W**	**WP**	**W**	**WP**	**W**	**WP**	**W**	**WP**
1	10.00	10.00	15.00	15.00	20.00	20.00	25.00	25.00	30.00	30.00
2	19.25	20.50	45.75	61.20	82.50	88.75	126.00	138.25	251.50	270.70
3	31.75	27.25	93.25	188.00	249.25	273.00	318.50	371.25	576.75	609.00
4	29.25	41.75	86.50	202.25	234.50	298.50	306.25	389.75	563.25	651.25
5	18.50	57.50	78.00	240.75	215.50	330.25	285.25	414.00	559.00	677.75
6	5.25	70.25	52.25	264.50	185.50	352.75	278.50	448.50	501.50	695.00
7	0.50	94.50	26.50	283.75	122.50	388.50	262.50	471.00	467.00	723.50
8	0.00	121.75	15.50	299.25	116.25	402.75	235.25	499.75	445.25	702.75
9	-	-	5.00	316.50	93.75	426.25	204.75	519.50	411.75	678.25
10	-	-	3.25	332.75	65.25	450.50	178.75	549.00	378.50	623.75
11	-	-	0.00	364.25	28.50	470.25	123.5	582.25	316.50	601.50
12	-	-	-	-	12.25	492.75	102.25	600.75	262.25	592.00
13	-	-	-	-	4.50	501.00	74.75	624.25	210.75	588.50
14	-	-	-	-	0.00	536.50	47.50	648.75	187.75	600.75
15	-	-	-	-	-	-	16.50	678.50	105.00	610.25
16	-	-	-	-	-	-	0.00	706.00	52.75	649.50
17	-	-	-	-	-	-	-	-	3.75	678.75
18	-	-	-	-	-	-	-	-	0.00	702.00

*Mean of 4 observations ; W- with predator; WP- without predator

Table 15. Population build up of *Neoseiulus longispinosus* as influenced by predator: prey ratio

Days	***Predator population at different prey densities (Number per leaf)**				
	10	**15**	**20**	**25**	**30**
1	1.00	1.00	1.00	1.00	1.00
2	3.00	4.00	3.50	4.00	4.00
3	6.25	7.25	9.25	9.00	9.25
4	9.50	9.75	12.25	12.50	12.50
5	12.25	13	15.5	16.25	15.5
6	9.50	16.25	19.75	22.50	26.50
7	4.00	20.25	25.75	29.5	32.75
8	2.25	18.25	29.50	34.50	39.50
9	-	16.75	33.25	39.25	43.25
10	-	15	37.00	42.25	47.75
11	-	4.25	26.75	49.50	54.50
12	-	-	16.25	56.75	58.75
13	-	-	15.5	62.25	61.25
14	-	-	5.25	39.50	66.50
15	-	-	-	14.75	73.00
16	-	-	-	7.50	42.25
17	-	-	-	-	17.00
18	-	-	-	-	8.50

*Mean of 4 observations

Table 16. Growth rate of *Tetranychus urticae* and *Neoseiulus longispinosus* as influenced by prey densities

Days	Growth rate of prey and predator (%) at different prey densities													
	10			**15**			**20**			**25**			**30**		
	Prey with predator	Prey alone	Predator	Prey with predator	Prey alone	Predator	Prey with predator	Prey alone	Predator	Prey with predator	Prey alone	Predator	Prey with predator	Prey alone	Predator
8	-100.00	21.90	37.83												
11				-100.00	31.10	36.33									
14							-100.00	23.47	28.11						
16										-100.00	19.65	24.61			
18													-100.00	18.13	20.13

4.4.3. Prey Stage Preference

The preference of both deutonymph and adult of *N. longispinosus*, to three different stages (egg, larva and deutonymph) of the prey, *T. urticae* was studied in the laboratory. The results revealed that both nymph and adult stage of the predator consumed more eggs than larva and deutonymph of the prey.

At a prey density of 5:5:5 (15) the predator deutonymph consumed 3.00 (68.66 %), 0.90 (20.83%) and 0.50 (10.50 %) egg, larva and deutonymph, respectively. The response of predator deutonymph to the higher prey densities of 10:10:10 and 15:15:15 were also similar. The deutonymph consumed 5.20 eggs (60.21 %), 2.20 larvae (25.01 %) and 1.3 deutonymphs (14.77 %) at a prey density of 10:10:10 (30) whereas, at a prey density of 15:15:15 (45) the prey consumption recorded by predator deutonymph was 5.30 (58.06 %) eggs, 2.50 (26.93 %) larvae and 1.40 (15.01 %) deutonymphs, respectively (Table 17).

Table 17. Preference of *Neoseiulus longispinosus* deutonymph towards different stages of *Tetranychus urticae*

Prey density	***Rate of predation**					
	5:5:5		**10:10:10**		**15:15:15**	
	Number	**%**	**Number**	**%**	**Number**	**%**
Egg	3.00	68.66[a] (8.31)	5.20	60.21[a] (7.79)	5.30	58.06[a] (7.65)
Larva	0.90	20.83[b] (4.61)	2.20	25.01[b] (5.05)	2.50	26.93[b] (5.23)
Nymph	0.5	10.50[b] (3.31)	1.3	14.77[c] (3.90)	1.4	15.01[c] (3.93)

*Mean of 10 observations

Figures followed by the same alphabets did not differ significantly (P=0.01)

Figures in parenthesis are square root transformed values

At a prey density of 5:5:5 (15), the adult *N. longispinosus* consumed 4.30 eggs (58.31 %), 1.90 larvae (24.15 %) and 1.40 deutonymphs (17.53 %). The consumption of egg, larva and nymph at a prey density of 10:10:10 (30) was 6.90 (51.28 %), 3.70 (25.67 %) and 3.20 (23.04 %) respectively, whereas, at a prey density of 15:15:15 (45) the predator adult consumed 8.20 eggs (58.03 %), 3.60 larvae (25.29 %) and 2.40 deutonymphs (16.61 %), respectively (Table 18).

Table 18. Preference of *Neoseiulus longispinosus* adult towards different stages of *Tetranychus urticae*

Prey density	***Rate of predation**					
	5:5:5		**10:10:10**		**15:15:15**	
	Number	**%**	**Number**	**%**	**Number**	**%**
Egg	4.30	58.31[a] (7.66)	6.90	51.28[a] (7.19)	8.20	58.03[a] (7.65)
Larva	1.90	24.15[b] (4.96)	3.70	25.67[b] (5.11)	3.60	25.29[b] (5.07)
Nymph	1.40	17.53[b] (4.24)	3.20	23.04[b] (4.85)	2.40	16.61[c] (4.13)

*Mean of 10 observations ** Figures in parenthesis are square root transformed values

Figures followed by the same alphabets did not differ significantly (P=0.01)
Figures in parenthesis are square root transformed values

4.5. ESTIMATION OF OPTIMUM PREDATOR: PREY RATIO

To identify the optimum predator prey ratio at which effective management of prey mites could be brought about, experiments were conducted both in the laboratory as well as in polyhouse using a range of predator: prey ratios.

4.5.1. Laboratory Evaluation of Optimum Predator: Prey Ratio

To estimate the optimum predator prey ratio of *N. longispinosus* to *T. urticae* in laboratory, the predator was released at seven different densities (1, 2, 3, 4,

5, 10 and 20) on cucumber leaves with hundred mixed stages of prey along with a control (without predator). The results are presented in Table 19.

Table 19. Influence of predator: prey ratio on the population of *Tetranychus urticae* and *Neoseiulus longispinosus* on cucumber (*In vitro* study)

Predator : prey ratio	**No. of predators released/ 100 prey**	***No. of prey**			***No. of predator**		
		3rd day	**7th day**	**10th day**	**3rd day**	**7th day**	**10th day**
1: 100	1	277.00^{b} (16.65)	398.67^{b} (19.97)	171.00^{b} (13.09)	8.33^{d} (2.97)	33.00^{cd} (5.78)	67.33^{a} (8.23)
1: 50	2	230.67^{b} (15.20)	182.67^{c} (13.53)	45.00^{c} (6.74)	9.00^{d} (3.08)	31.67^{cd} (5.67)	35.33^{d} (5.98)
1: 33	3	282.67^{b} (16.82)	148.00^{cd} (12.18)	17.33^{d} (4.22)	26.00^{c} (5.14)	45.00^{c} (6.74)	41.33^{c} (6.46)
1: 25	4	243.66^{b} (15.62)	161.66^{cd} (12.73)	6.33^{d} (2.61)	23.67^{c} (4.91)	68.33^{b} (8.29)	47.33^{b} (6.91)
1: 20	5	262.33b^{b} (16.21)	127.33^{d} (11.30)	2.66^{d} (1.77)	34.67^{b} (5.93)	85.33^{a} (9.26)	67.00^{a} (8.21)
1: 10	10	49.00^{c} (7.03)	1.33^{e} (1.35)	0.00^{d} (0.70)	23.67^{c} (4.91)	39.33^{c} (6.31)	23.66^{e} (4.91)
1: 5	20	5.67^{c} (2.48)	0.00^{e} (0.70)	0.00^{d} (0.70)	96.33^{a} (9.84)	0.00 (0.70)	0.00 (0.70)
Control		609.00^{a} (24.68)	756.00^{a} (27.50)	561.00^{a} (16.65)	-	-	-
CD (0.05)		78.88	38.25	25.91	5.01	8.55	4.85

*Mean of three observation

Figures followed by the same alphabets did not differ significantly (P=0.01)

Figures in parenthesis are square root transformed values

Three days after the release of the predator, the mean number of prey mites recorded were 277.00, 230.67, 282.67, 243.66, 262.33, 49.00 and 5.67 at predator densities of 1, 2, 3, 4, 5, 10 and 20, respectively where as in the control 609.00 prey mites were recorded on the third day. On third day after the release of the predatory mites 8.33, 9.00, 26.00, 23.67, 34.67, 23.67 and 96.33 preadtory mites were recorded at prey densities of 1, 2, 3, 4, 5, 10 and 20, respectively.

On seventh day, 398.67, 182.67, 148.00, 161.66, 127.33 and 1.33 prey mites were recorded at predator densities of 1, 2, 3, 4, 5 and 10, respectively. The predatory mite count recorded on the same day was 33.00, 31.67, 45.00, 68.33, 85.33 and 39.33, respectively. At a release rate of 20 predators for 100 prey mites, no prey and predator were recorded in the experimental arena on the seventh day, where as in control, 756.00 prey mites were recorded on the seventh day.

On tenth day, a drastic reduction in the number of prey mites was recorded in all the treatments. At predator densities of 1, 2, 3, 4 and 5, prey mite counts recorded were 171.00, 45.00, 17.33, 6.33 and 2.66, respectively. However at densities of 10 and 20, prey mites were zero. In control, 561.00 prey mites were recorded on tenth day. The predators recorded a population of 67.33, 35.33, 41.33, 47.33, 67.00 and 23.66 at densities of 1, 2, 3, 4, 5 and 10, respectively.

4.5.2. Standardization of Optimum Predator: Prey Ratio in Polyhouse

To optimize the optimum predator: prey ratio of *N. longispinosus* to *T. urticae* required for effective suppression of spider mites in polyhouse conditions, an experiment was laid out by releasing predators at four different predator: prey ratios of 1: 20, 1: 25, 1: 33 and 1: 50 on cucumber as host plant. The population of *T. urticae* on cucumber on 5, 10 and 15 days after first and second release of the predator is furnished in Table 20.

The prey counts at five days after the first release of the predator were significantly lower at the ratios of 1: 20 (1.86 mites/cm^2) and 1: 25 (1.91 mites/cm^2) which were on par with each other. However, the prey population recorded at 1: 33 (4.21 mites/cm^2) was significantly higher compared to 1: 50 (3.34 mites/cm^2). In the control, an average of 8.24 prey mites per cm^2 was recorded.

Table 20. Influence of predator: prey ratio on the population of *Tetranychus urticae* on cucumber in polyhouse after the release of predator

Predator: prey ratio (No. of predators/ 100 prey)	*No. of prey mites/ cm^2					
	First release			Second release		
	5DAR	10 DAR	15 DAR	5DAR	10 DAR	15 DAR
1:20 (5)	1.86^d (1.53)	3.36^c (1.96)	4.99^d (2.34)	3.53^d (2.00)	2.32^d (1.67)	1.03^d (1.23)
1:25 (4)	1.91^d (1.55)	3.31^c (1.95)	5.61^d (2.47)	3.96^d (2.11)	2.38^d (1.69)	1.11^d (1.26)
1:33 (3)	4.21^b (2.17)	4.83^b (2.30)	11.12^c (3.40)	6.93^c (2.72)	8.33^c (2.97)	7.14^c (2.76)
1:50 (2)	3.34^c (1.95)	4.81^b (2.30)	12.05^b (3.54)	7.67^b (2.85)	11.37^b (3.44)	9.92^b (3.22)
Control (0)	8.24^a (2.95)	12.89^a (3.65)	13.86^a (3.78)	15.36^a (3.98)	19.79^a (4.50)	20.60^a (4.59)
CD	0.44	0.82	0.88	0.58	0.54	0.60

*Mean of 45 observations (All life stages)

DAR –Days After Release

Figures followed by the same alphabets did not differ significantly (P=0.01)

Figures in parenthesis are square root transformed values

Ten days after the first release of the predator, significantly lower prey mite population was recorded at the ratios of 1: 20 (3.36 mites/cm^2) and 1: 25 (3.31 mites/cm^2) which were on par with each other. On this day, the prey mite count at the ratios 1: 33 (4.83 mites/cm^2) and 1: 50 (4.81 mites/cm^2) were on par with each other. The prey mite count recorded in the control was 12.89 mites per cm^2 leaf area which was significantly higher than all other treatments.

On fifteenth day after the first release of the predator, 4.99 and 5.61 prey mites were recorded per cm^2 leaf area at the ratios 1:20 and 1:25 which were on par with each other. The ratio of 1: 33 (11.12 mites/cm^2) recorded significantly higher prey mite population compared to the ratio 1: 50 (12.05 mites/cm^2). However, all these ratios were significantly superior to the control (13.86 mites/cm^2) in reducing prey the mite population.

Five days after the second release of the predator, 3.53 and 3.96 prey mites were recorded per cm^2 leaf area at ratios of 1: 20 and 1: 25, respectively. These ratios were found to be significantly superior to other treatments and were on par with each other. At ratios of 1: 33 and 1: 50, 6.93 and 7.67 prey mites were recorded per cm^2 leaf area, respectively. Significantly lower prey population was recorded at the ratio 1: 33 compared to the ratio of 1: 50. However the control treatment recorded a significantly higher prey count of 15.36 mites per cm^2 leaf area.

Ten days after the second release of the predator, significantly lower prey mite populations were recorded in 1: 20 (2.32 mites/ cm^2) and 1: 25 (2.38 mites/ cm^2). This was followed by the ratios of 1: 33 (8.33 mites/ cm^2) and 1: 50 (11.37 mites/ cm^2) which differed from each other significantly. In the control, 19.79 mites per cm^2 leaf area were recorded.

Significant reduction in prey mite population was recorded fifteen days after the second release of the predator with a record of 1.03 and 1.11 mites per cm^2 leaf

area at ratios of 1: 20 and 1: 25, respectively. These ratios were found to be significantly superior to other ratios and were on par with each other. Fifteen days after the second release, 7.14 prey mites/ cm^2 was recorded in the ratio 1: 33 which was superior to the mite count at 1: 50 (9.92 mites/ cm^2). In the control, significantly higher mite count of 20.60 was recorded.

Discussion

5. DISCUSSION

The observations and inferences on the investigations carried out on the incidence, crop phenology relationship, natural enemies and biology of the two spotted spider mite, *Tetranychus urticae* Koch and biology, efficacy, prey stage preference and optimum predator: prey ratio of *Neoseiulus longispinosus* (Evans) for biological control of *T. urticae* in polyhouse are discussed below in the light of available literature.

5.1. INCIDENCE, CROP PHENOLOGY RELATIONSHIP AND NATURAL ENEMIES OF *Tetranychus urticae*

5.1.1. Diversity of Spider Mites

The survey on the diversity of mite fauna in the polyhouses covering four districts of Kerala recorded five species of phytophagous mites belonging to two different families on cucumber. The phytophagous mites recorded in the present study included, the spider mite *T. urticae, Tetranychus truncatus* Ehara*, Tetranychus okinawanus* Ehara, *Eutetranychus orientalis* (Klein) and the yellow mite *Polyphagotarsonemus latus* (Banks). Spider mites belonging to the family Tetranychidae were found to be the predominant phytophagous mites on cucumber. With the booming of polyhouse cultivation of vegetable crops in Kerala, mites have emerged as a serious pest of vegetables. However, studies on the species diversity of mites infesting cucumber in Kerala are limited. Sudharma (1996) reported *T. urticae* (= *T. cinnabarinus*) as a pest of field grown cucumber from Thiruvananthapuram district of Kerala. *T. urtiae* was also reported as the dominant species of mite pest on vegetable crops (Binisha and Bhaskar, 2013) and as the most important mite pest of okra (Krishna and Bhaskar, 2014) in Thrissur district, Kerala.

In the present study, *T. truncatus* was found to be the predominant species of mite on cucumber which was recorded from all the polyhouses surveyed. It was

reported as an emerging and potential pest of mulberry from Karnataka by Srinivasa *et al.* (2012). Bennur *et al.* (2015) has recently reported *T. truncatus* as the dominant mite pest of vegetables in Thrissur district of Kerala. This is the first record of *T. truncatus* on cucumber in polyhouse from India.

Tetranychus okinawanus was first reported from Kerala on adenium during 2013 (AINPAA, 2015). Later Bennur *et al*. (2015) reported *T. okinawanus* on cucumber for the first time from Kerala. The species was first reported and described from Ryukyu islands of Okinawa, Japan (Ehara, 1995) on *Pueraria lobata* (Willd.).

The two recently recorded species on cucumber observed during the present study *viz. T. truncatus* and *T. okinawanus* were confirmed by molecular characterization. The findings matched with that of Bennur *et al.* (2015) on vegetable crops from Kerala.

5.1.2. Incidence and Crop Phenology Relationship of *Tetranychus urticae* to Cucumber

Though the mite incidence was recorded through out the cropping period on cucumber, significantly higher population was recorded during late fruiting and early vegetative stage (Fig.1). Studies relating to the relationship between mite infestation and stage of the crop are scarce, especially on cucumber. However, similar studies on other crops show that the peak incidence varies at different stages of crop growth. Higher population during later stage of crop has been documented in several cases. Jayasinghe and Mallik (2010), for instance, reported that middle stage of the tomato crop was the most critical period for mite infestation. Alatawi *et al*. (2007) reported that older *Impatiens* sp. exhibited greater damage than younger plants. Nandini *et al*. (2012) observed that significantly lower mite infestation was recorded during young stage (60 days) of bell pepper plants. The peak infestation was recorded during the later stages of the crop, *i.e* 180 days. She also noted that the incidence of mites increased

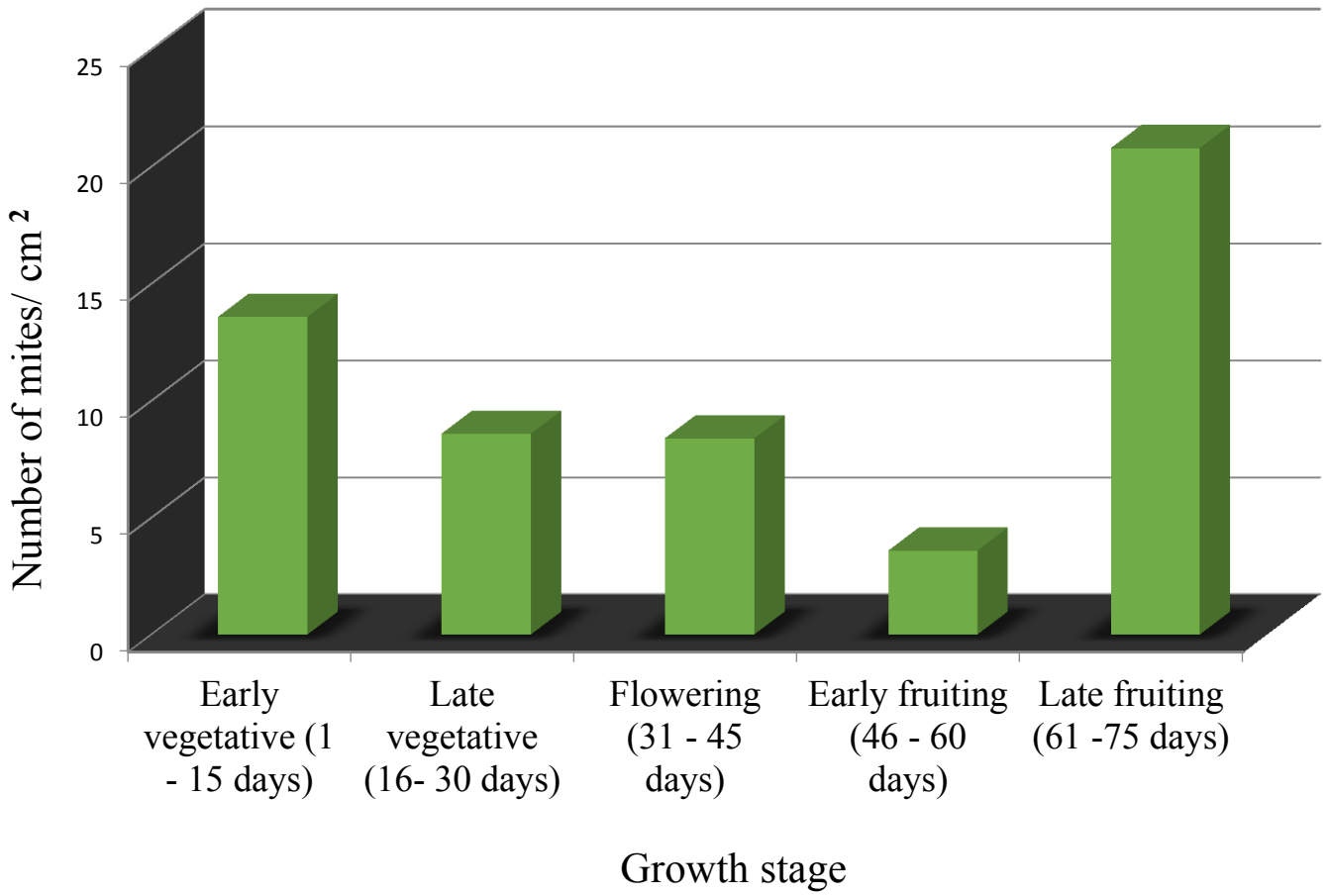


Fig. 1. Population of spider mites as influenced by growth stages of cucumber

gradually with the advancement of crop age. As the crop advanced in age, it gave more time for mite pests to multiply in numbers besides allowing itself to put up more foliage and food.

5.1.3. Natural Enemies of Spider Mites

5.1.3.1. Insect Predators of Spider Mites

Four species of predatory insects were recorded on spider mites on cucumber in polyhouse during the study. Predatory insects recorded were *Stethorus pauperculus* (Weise) (Coleoptera: Coccinellidae), *Oligota* sp. (Coleoptera: Staphylinidae), *Scolothrips* sp. (Thysanoptera: Thripidae) and an unidentified species of Cecidomyiidae. These insects were reported as major predators of spider mites on various crops by different authors.

The important species of *Stethorus* reported as specific predators of mites are *Stethorus pauperculus* Weise (Kapur, 1948), *S. tetranychi* Kapur (Puttarudriah and Channabasavanna, 1955), *S. keralicus* Kapur (Puttaswamy and Rangaswamy, 1976), *Parastethorus histrio* (Chazeau) (Dhooria, 1981), *S. parcepunctatus* Kapur (Gupta, 2001) and *S. gilvifrons* (Mulsant) (Sarmah and Bhattacharya, 2002).

Insect predators like *Oligota* sp., *Scolothrips rhagebianus* Preiesner and two species of predatory bugs belonging to the family Anthocoridae and Miridae were observed feeding on tetranychid mites of brinjal in Dharward district of Karnataka (Prasanna and Kumar, 2008).

The predatory beetles, *Oligota pygmaea* and *S. gilvifrons* were reported to be efficient predators of *Olygonychus coffeae* in tea. Attempts were made on the mass release of the same in organic tea fields (Perumalsamy *et al*., 2010). *Oligota* sp. and *S. pauperculus* are the most widely seen coccinellid predators of *T. urticae* on okra in

Tamil Nadu and have been evaluated as biocontrol agents. The predator prey ratio of 5: 150 was found to be the ideal for both the beetle predators (Jeyarani *et al.*, 2012).

5.1.3.2. Fungal Pathogens of Spider Mites

During the study, mycosis of spider mites was observed in a polyhouse of Thrissur district. A fungal pathogen *Acremonium strictum* W Gams. was isolated from the mycosed mites of cucumber. This is a new report of *A. strictum* as a pathogen on mites. Recently, *Acremonium zeylanicum* (Petch) W. Gams, *Neozygetus floridana*, and *Conidiobolus* sp. were isolated from mycosed mites on bhindi in polyhouse of Thrissur district (Krishna *et al*., 2014).

Several other fungi have also been reported as efficient pathogens of spider mites. Natural infection of *Beauveria bassiana* on tetranychid mites were reported on tomato, beans and okra from Karnataka during 2004- 2005 (Kalmath *et al*., 2007).

Cladosporium cladosporoides (Frensen.) was reported to cause 75 to 95 per cent natural mortality of spider mites on cowpea and okra crops in Coimbatore (AINPAA, 2011).

5.1.3.3. Predatory Mites of Spider Mites

Six species of predatory mites belonging to five families were recorded from cucumber in polyhouse. These were *N. longispinosus*, *Agistemus garrulous* Chaudhari*, Tydeus gossabaensis* Gupta*, Amblyseius paraaerialis* (Muma)*, Cunaxa* sp. and *Cheyletus* sp. Karmakar and Gupta (2010) reported *Amblyseius* (=*Neoseiulus*) *longispinosus, Amblyseius largoensis* (Muma) and *Agistemus* sp. as the dominant and efficient predators of spider mites from major parts of India.

Sheeja (2010) conducted a survey on the phytoseiid mites and associated pests were carried out in two districts of Kerala, Kozhikode and Malappuram. Among the 18 species of phytoseiid mites identified, five species *viz., A. largoensis*,

Typhlodromips suknaensis Gupta, *Amblyseius guptai* sp. nov.*, Paraphytoseius multidentatus* Swirski and Shechter, and *Phytoseius rachelae S*wirski and Shechter were recognized as the most common and abundant phytoseiid predators in the region. Of these, *A. largoensis*, *T. suknaensis* and *A. guptai* had broad range of feeding habits. On the other hand, *P. multidentatus* and *P. rachelae* showed more preference to insect pests than mite pests.

Study on the relative abundance of predatory mites associated with phytophagous mites in different vegetable crops revealed the occurrence of 15 species of predatory mites belonging to six genera *viz*., *Amblyseius*, *Typhlodromips*, *Neoseiulus*, *Euseius*, *Phytoseius* and *Paraphytoseius* under the suborder Mesostigmata. In the genus *Amblyseius,* eight species were recorded *viz*., *Amblyseius aerialis, Amblyseius indirae, Amblyseius channabasavannai, Amblyseius kundurukkae, Amblyseius largoensis, Amblyseius orientalis, Amblyseius herbicolus* and *Amblyseius kulini.* Two species recorded in the genus *Euseius* were, *E. coccinea* and *E. alstoniae*. *T. syzygii* was the only species recovered from the genus *Typhlodromips.* The genus *Phytoseius* included the species, *P. wainsteini* and *P. punjabensis*. A single species was obtained from genus *Neoseiulus, viz*., *N. longispinosis. P. scleroticus* was the only species in the genus *Paraphytoseius* (Haneef and Sadanandan, 2013).

Binisha and Bhaskar (2013) reported Phytoseiidae as the dominant predatory mite family associated with vegetables of Thrissur district of Kerala. They reported seven species of Phytoseiids, namely, *A. paraaerialis*, *P. orientalis* Narayanan, *N. longispinosus*, *Phytoseius* sp. *Euseius macrospatulatus* Gupta, *Typhlodromips* sp. and *Scapulaseius* sp. associated with spider mites.

Similar reports were also made by Maheswary (2015) confirming the dominance of the family Phytoseiidae as mite predators in vegetables. Nine species of Phytoseiid predators recorded in her study were *N. longispinosus, A. paraaerialis, A.*

largoensis, E. macrospatulatus, Euseius sp. nr. *prasadi, Typhlodromips syzygii, P. orientalis, Phytoseius intermedius* and *Scapulaseius sp.*. Other major predatory mites recorded were Cunaxa sp. (Cunaxidae), *Bdella khasyana* (Bdellidae), four species of the family Stigmaeidae *viz., Agistemus gamblei, A. fleschneri, A. garrulus* and *A. macrommatus* and two species of Tydeidae *viz., Tydeus gossabaensis* and *Pronematus anconai. Euseius* sp. nr. *prasadi, P. intermedius*, *B. khasyana*, *A. fleschneri, A. garrulous, A. macrommatus*, *T. gossabaensis* and *P. anconai* were reported for the first time from Kerala.

5.2. BIOLOGY OF *Tetranychus urticae* ON CUCUMBER

Studies on the biology of *T. urticae* were conducted in the Acarology laboratory at 27.05 ±0.68ºC and 90.12 ± 6.88 per cent relative humidity during June-July, 2013 on leaf discs of cucumber. The developmental stages of *T. urticae* on cucumber consisted of egg, larva, protonymph, deutonymph and adult, as reported by Krishna and Bhaskar (2014) on okra in Thrissur district. The quiescent intervals such as nymphochrysalis, deutochrysalis and teleiochrysalis recorded in the study have been reported by Riahi *et al*. (2011) in *T. urticae* infesting peach as also by Krishna and Bhaskar (2014) in *T. urticae* infesting okra.

The development of egg, larva, protonymph and deutonymph of *T. urticae* on cucumber was 2.48, 0.70, 0.52, 0.77 days for females and 2.85, 0.56, 0.57, 0.71 days for males, respectively. The total developmental period of *T. urticae* on cucumber was 6.75 days for males and 7.15 days for females. Several workers had already reported a similar trend in the developmental biology of different species of *Tetranychus* infesting various crops. Krishna and Bhaskar (2014) observed that the males of *T. urticae* completed life cycle in 6.73 days on okra where as female completed their life cycle in 7.52 days at 30ºC. Males of *T. urticae* completed the life cycle in 6.3 days and females in 6.5 days (El- Wahed and El- Halawany, 2012).

Rajkumar (2003) also recorded a shorter duration of development for male (10.70) compared to female (12.36 days) in *T. urticae* on jasmine.

The deutonymphs and adults of female *T. urticae* developed on cucumber were larger in size compared to males as already reported by Krishna and Bhaskar (2014) in *T. urticae* on bhindi. Adult male was reddish green or light red in colour with the body tapering postereorly to a blunt point, whereas the females were larger, plumpy and dark red in colour with longer setae all over the body. Similar sexual dimorphism was also reported by Seeman and Beard (2011) in *T. urticae* as well as Krishna and Bhaskar (2014) in *T. urticae*.

Tetranychus urticae showed a general preference to lay eggs on the undersurface of the leaf, near the veins and midrib of cucumber which was in agreement with the findings of Krishna and Bhaskar (2014). 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: 4.6. Female biased sex ratio (1: 5.8) was also reported in *T. urticae* on okra by Krishna and Bhaskar (2014). Kaimal and Ramani (2011) also 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 mate with several females (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, even though the proportion of males in a population remains low.

The progenies of unmated females of *T. urticae* were only males. This is in conformity with earlier reports by Sabelis (1981) and Krishna and Bhaskar (2014) in *T. urticae*. Arrhenotoky is also reported in other species of *Tetranychus* as well (Manjunatha and Puttaswamy, 1989; Bonato and Gutierrez, 1999).

The fecundity of mated and unmated females of *T. urticae* in cucumber was low compared to that on bhindi as reported by Krishna and Bhaskar (2014). Mated female and unmated females laid only 47.91 and 36.08 eggs, on cucumber respectively as against the report by Krishna and Bhaskar (2014) who reported as many as 108 eggs by mated female and 77 eggs by unmated female on bhindi. High fecundity of mated females of *T. urticae* was also reported by Rajkumar (2003). The study on biology of *T. urticae* on cucumber was carried out at a temperature of 27.05 ±0.68ºC and 90.12 ±6.88 % relative humidity while the study on *T. urticae* on bhindi by Krishna and Bhaskar (2014) was carried out at a temperature of 30ºC. 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^{o}C (El-Wahed and El-Halawany, 2012). The low temperature during the study period might be the reason for recording a low fecundity of 47.91 eggs at 27^{o}C in the present study. The higher egg viability of 92.55 per cent observed in the current study, is an evidence of the high biotic potential of this species. The egg viability of 92.10 per cent in *T. urticae* was recorded on okra by Krishna and Bhaskar (2014), while Silva *et al*. (2009) reported a higher egg viability of 96.5 per cent in *T. urticae* on gerbera.

In the present study, it was found that males lived longer than females as already reported by Krishna and Bhaskar (2014). Similar findings were recorded by Puttaswamy and Channabasavanna (1980) in *T. ludeni* also.

The present study was carried out in laboratory at a temperature of 27 ºC. however the temperature inside polyhouses in Kerala are on a higher range favouring the population build up of mites, where it is expected to develop at a faster rate than that was recorded in the laboratory at a lower temperature. At a temperature of 30 ºC, the life cycle of *T. urticae* would be short and the fecundity would be high as reported by Krishna and Bhaskar (2014).

5.3. BIOLOGY OF *Neoseiulus longispinosus* ON *Tetranychus urticae*

The studies on the biology of *N. longispinosus* on *T. urticae* were conducted at 28.05 ± 0.90 ºC and 79.68 ± 8.11 per cent relative humidity during December-January, 2014. The developmental stages of *N. longispinosus* consisted of egg, larva, protonymph, deutonymph and adult. The studies on biology of *N. longispinosus* on *Polyphagotarsonemus latus* (Kongjarean, 2006) and *Oligonychus coffeae* (Rahman *et al*., 2012 b) recorded similar life stages in the development of the phytoseiid predator.

The embryonic developmental period was longer for female (1.57 days) than for male (1.46 days). The result is in agreement with the findings of Rahman *et al.* (2012 b). However, Ibrahim and Palacio (1994) observed longer incubation period for males compared to females. The larval, protonymphal and deutonymphal periods observed in the present study is similar to the reports of Ibrahim and Palacio (1994) and Rahman *et al*. (2012 b). In the present study, the larva of *N. longispinosus* was found to be a non feeding stage which was in accordance with the findings of Ibrahim and Palacio (1994). However, Rahman *et al.* (2012 a) reported that larvae of *N. longispinosus* fed on eggs, protonymph and deutonymph of *O. coffeae*.

Males were smaller and elongate than females. The male body size of *Neoseiulus californicus* and *Phytoseiulus persimilis* were smaller compared to that of female as reported by Walzer and Schausberger (2014). They also observed that large female body size has advantage in fecundity and mating behavior.

In the present study, the total developmental period from egg to adult was 3.91 days for male and 4.27 days for female at a temperature of 28ºC (Fig. 2). This is similar to the reports of developmental duration of *N. longispinosus* on *O. coffeae* by Rahman *et al.* (2012 b). On *O. coffeae* the predator completed its development in 4.0 and 4.2 days, respectively for males and females. Similar duration of development of *N. longispinosus* has been reported by Ibrahim and Palacio (1994) and Puspitarini (2010).

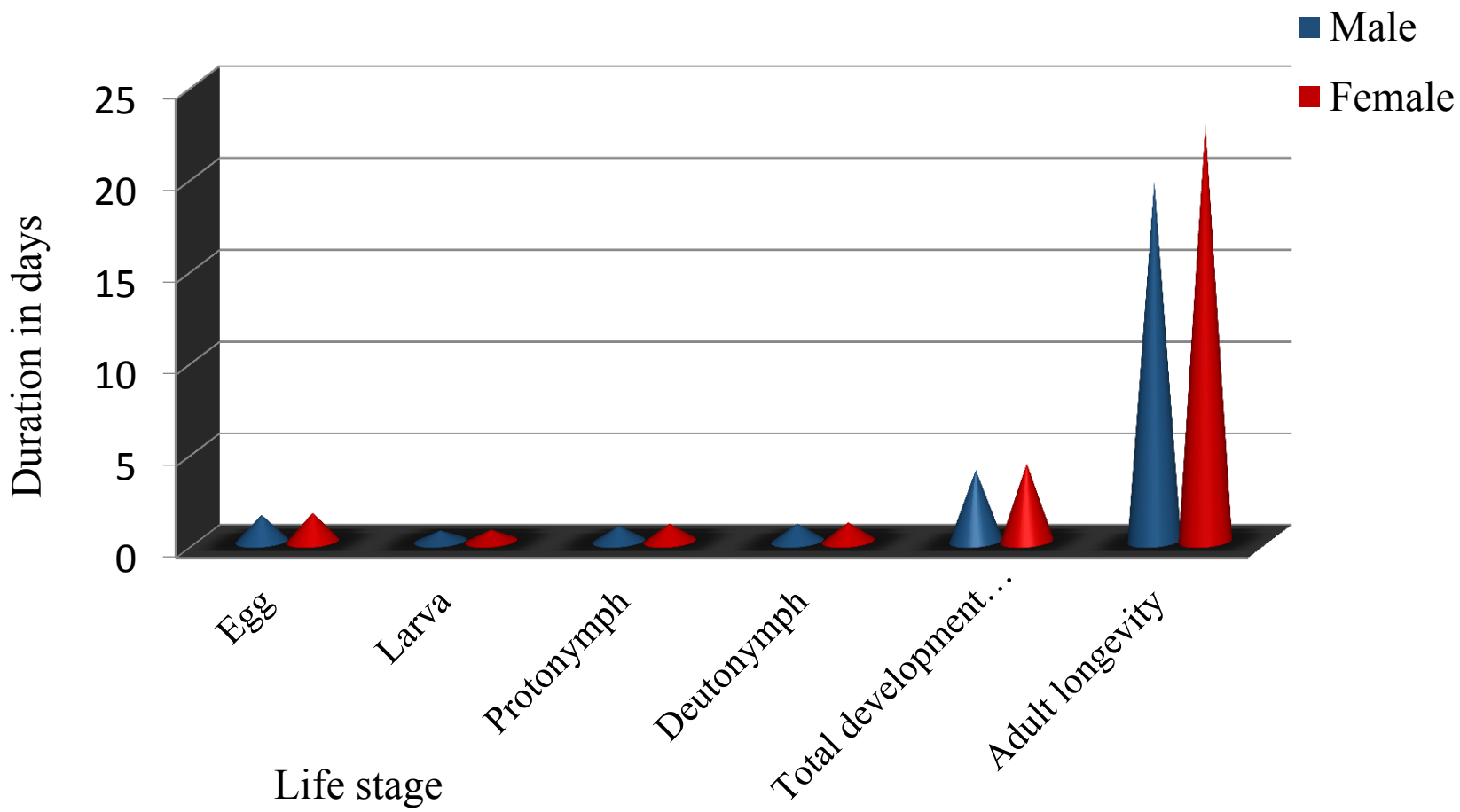


Fig. 2. Duration of developmental stages and adult longevity of *Neoseiulus longispinosus* on *Tetranychus urticae*

The shorter longevity of male (19.66 days) compared to female (22.75 days) (Fig. 2), recorded in the present study is in agreement with the findings of Puspitarini (2010) 13.95 days in males than that of female 15.42 days in females. However, Ibrahim and Palacio (1994) and Rahman *et al*. (2012 b) observed longer life for male than female. The study showed that the adult predator lived longer compared to the adult of prey, *T. urticae*.

The male mated immediately after moulting of the adult female and female mated more than once. The observation made in the mating behavior of *N. longispinosus* was similar to that reported by Ibrahim and Palacio (1994). They observed that mating occurred in female on the same day of emergence and was repeated several times during the reproductive life. Rahman *et al*. (2012 b) reported that adult female mated on the first day of emergence and started laying eggs the next day. Amano and Chant (1977) stated that phytoseiid mites require multiple mating to attain their full reproductive potential.

The sex ratio of *N. longispinosus* was found to be 1: 3.31. Female biased sex ratio was also reported by Ibrahim and Palacio (1994) and Puspitarini (2010) in *N. longsipsinosus* while Rahman *et al*. (2012 b) recorded a narrow male: female ratio of 0.47: 1.

The mean pre- oviposition, oviposition and post- oviposition periods recorded in the study for mated female was 1.97, 19.91 and 0.99 days, respectively. According to Ibrahim and Palacio (1994) the mated female started laying egg on the second day of adult emergence. The pre- oviposition, oviposition and post- oviposition periods of 1.0, 23.4 and 1.9 days, respectively were reported for *N. longispinosus* when fed on *O. coffeae* on tea (Rahman *et al*., 2012 b).

Though *N. longispinosus* recorded a fecundity of 31.33 eggs per female in the present study, Ibrahim and Palacio (1994) as well as Rahman *et al*. (2012 b) had reported a higher fecundity of 50.7 and 60.2 eggs per female, respectively. However,

Puspitarini (2010) reported a lower fecundity of 25.90 in *Amblyseius* (=*Neoseiulus*) *longispinosus* on *T. kanzawai*.

In the present study, the percent viability of eggs was only 89.30. But Ibrahim and Palacio (1994) reported egg hatchability of 99.5 per cent in *N. longispinosus*. However, at 30 ºC, 97 per cent egg hatchability was recorded in a study by Rahman *et al*. (2012 b).

The total development period of the predatory mite *N. longispinosus* was as short as 4.2 days compared to 7.15 days of prey mite *T. urticae* indicating that the predator would be able to complete almost two generations when the prey completes one. The study also showed that the adult female lived longer for 22.75 days compared to 13.04 days of female *T. urticae*. Shorter life cycle and longer adult longevity (Fig. 3) would favorably influence the efficiency of *N. longispinosus* as a predator of spider mites.

5.4. EFFICACY AND PREY STAGE PREFERENCE OF *Neoseiulus longispinosus*

5.4.1. Predatory Potential of *Neoseiulus longispinsous*

Studies on the predatory potential of *N. longispinsous* on *T. urticae* showed a positive correlation between rate of predation and prey density. However, the predation was found to get leveled off at a density of 40 (Figs. 4 and 5). At higher prey density, it is expected that the predator would spend lesser time for searching the prey and also chances of encounter with the prey would be higher. Hence the predator would feed at a faster rate which would lead to satiation point. This has been well explained by Canlas *et al*. (2006) in the case of *N. californicus* which exhibited increased rate of predation with increase in the prey density till a prey density of 40- 50. Ibrahim and Rahman (1997) also reported the plateauing of predatory potential at a prey density of 40.

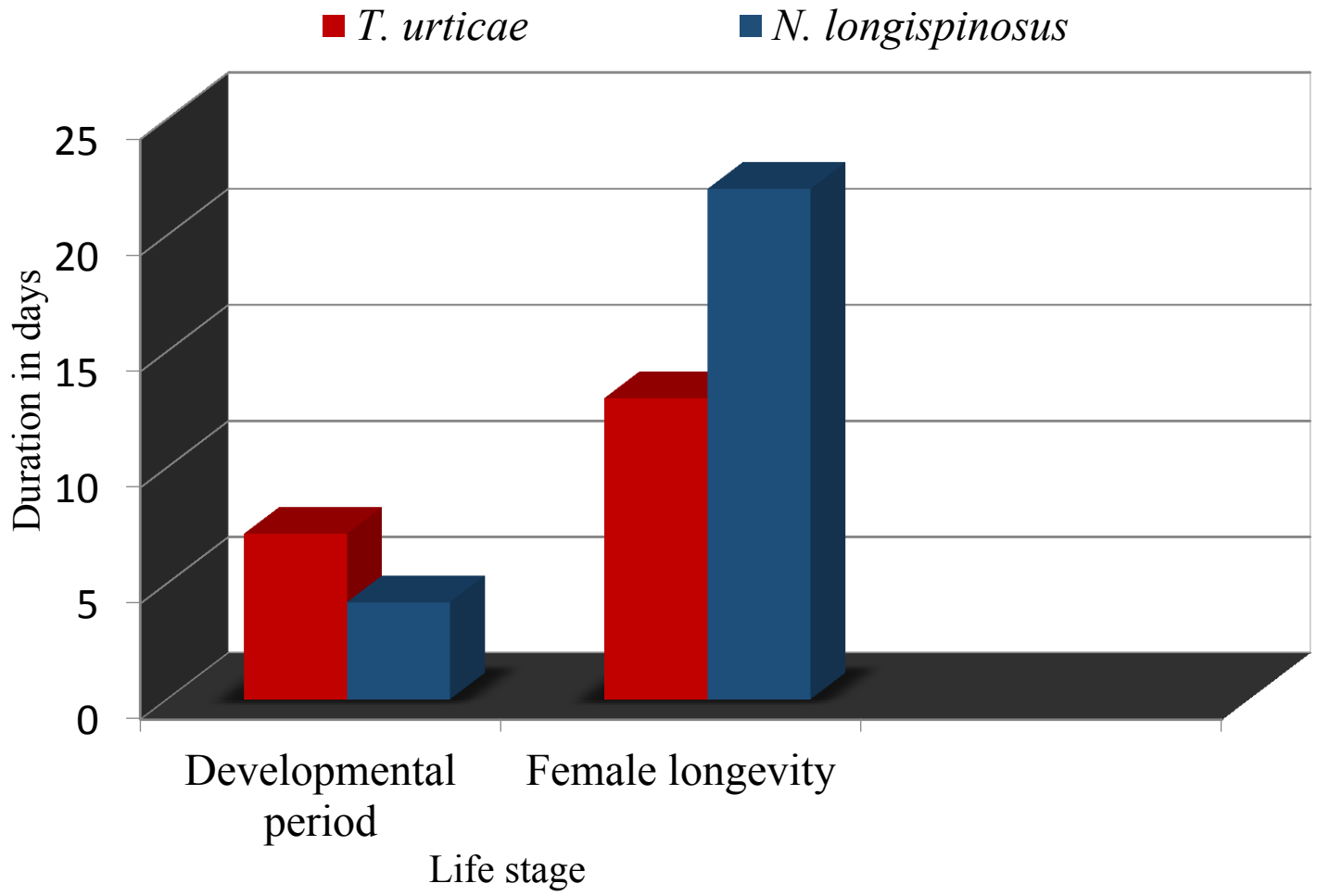


Fig. 3. Comparative biology of *Tetranychus urticae* and *Neoseiulus longispinosus*

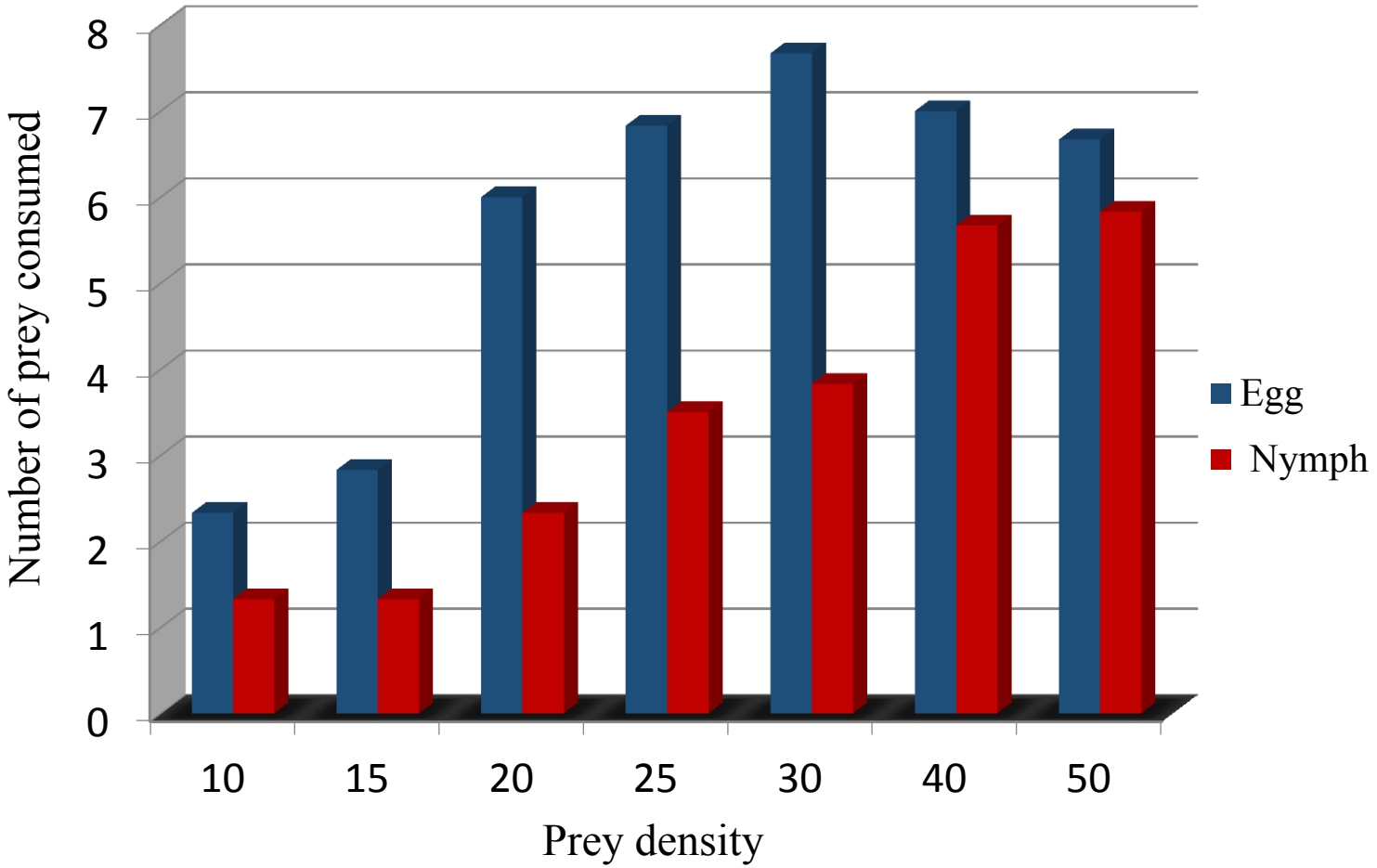


Fig. 4. Predatory potential of *Neoseiulus longispinosus* nymph at different prey densities

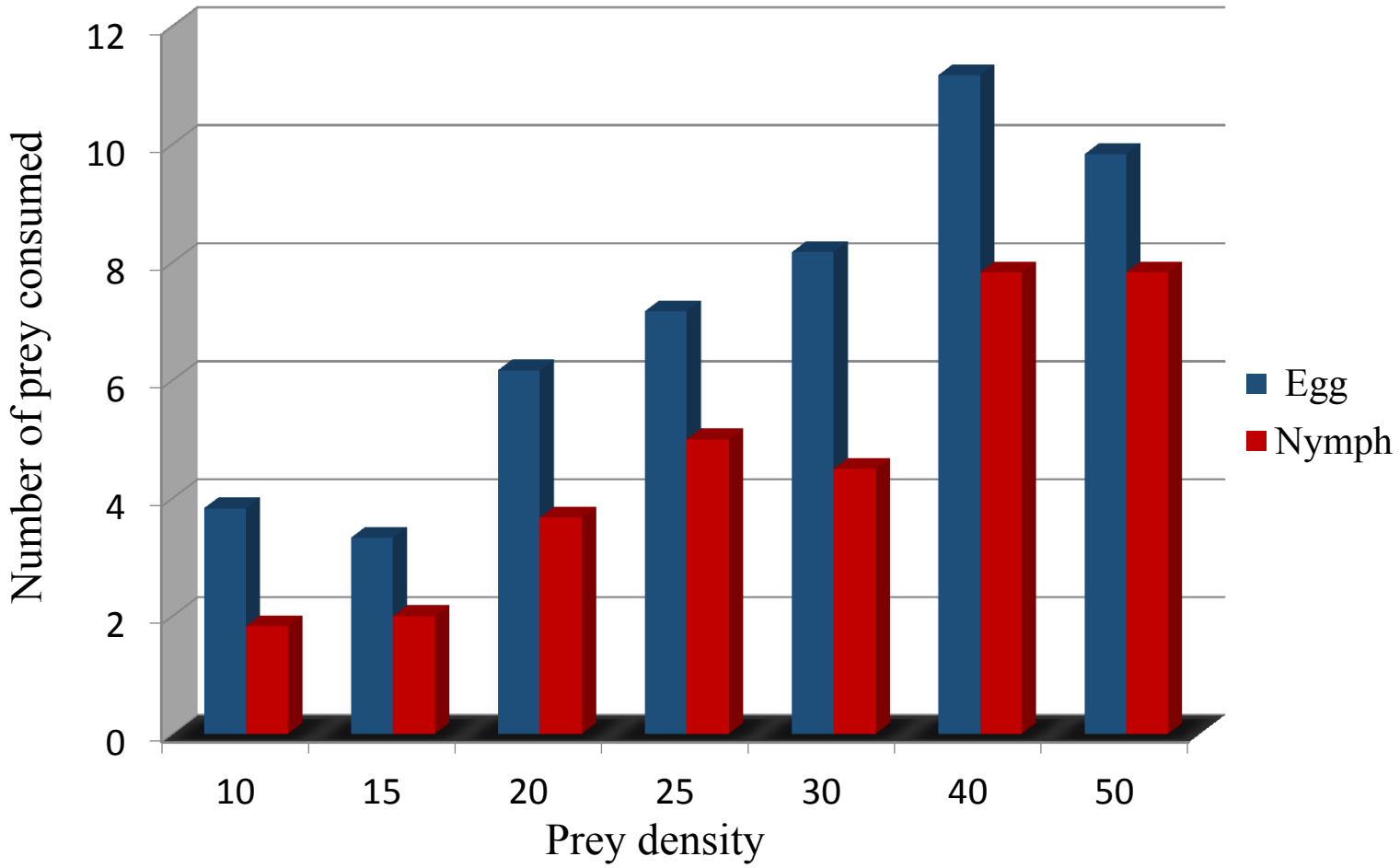


Fig. 5. Predatory potential of *Neoseiulus longispinosus* adult at different prey densities

In the present study, gravid female of *N. longispinosus* showed more predatory potential compared to the nymphal stage. One of the reasons that can be attributed for higher predation in the adult may be the bigger size of the body which in turn would demand more energy for the metabolic requirements. In addition, the major share of energy assimilated is required to be diverted for oogenesis in a mated female. This could be the reason for the increased predation exhibited by adult gravid female in the present study. Several authors have reported adult as the more potent stage than the nymph. Regardless of the stages of the prey, the gravid female was reported to maintain a higher rate of consumption over other stages (Ibrahim and Rahman, 1997; Rahman *et al.,* 2012 a).

In the present study, both nymph and adult exhibited higher predation on eggs compared to active stages of the prey at all densities tested. Studies of Ibrahim and Rahman (1997) and Rahman *et al*. (2012 a) also reported the same trend. As egg is an immobile stage, it is natural that the handling time taken by the predator on this would be shorter. In contrast, as the active stages resist the attack or try to evade the predator, it would become necessary for the predators to spend more time in handling the prey. This explains why the predator feeds more on eggs than active stages of *T. urticae*.

5.4.2. Time Needed to Control the Prey

In the present investigation, the study conducted to estimate the time needed to control the prey population showed that, that the time needed to eliminate the prey increased with increase in prey density. A single gravid female predator could control the mite population in eight days when released at an initial prey density of ten (1: 10) (gravid females of *T. urticae*). But, at a higher initial prey density of 30 (1: 30), the predator took 18 days for completely eliminating the prey population. A study conducted at Bangaluru, in tomato showed that at a ratio of 1: 50, predators required five weeks to eliminate the spider mites completely from the tomato plants, while at

release ratio of 1: 100, predator eliminated the spider mites in six weeks. At ratios of 1: 200 and 1: 400, predators failed to eliminate the spider mites completely during the study period. The results of the study clearly indicated that the predator would effectively control the prey population much earlier at a narrow predator: prey ratio than at wider ratios, as reported by Perumalsamy *et al.* (2010) and Rahman *et al.* (2012 a).

When the growth rate of prey was computed for different prey densities with and without predator, it was found that there was a substantial increase in growth of prey in the absence of predator. The growth rate of prey was found to decrease and that of predator was found to increase (Figs. 6 and 7). As the study was conducted with gravid females of prey and predator, it is expected that both prey and predator laid eggs leading to population build up (Fig. 8). However, the population build up of the predator is expected to be faster compared to that of prey due to shorter developmental duration of the predator as revealed in the present study on biology of *N. longispinosus*. This combined with the extended longevity of adults which are more potent as predators would effectively bring down the population of prey. These qualities of predator might have helped in reducing the growth rate of the prey significantly on release of the predator as against increased growth rate of prey in the absence of the predator.

5.4.3. Prey Stage Preference

The results of the study showed that both nymph and adult predators preferred egg stage of the prey compared to larva and nymph (Figs. 9 and 10). Mallik *et al.* (1998) observed that *N. longispinosus* consumed more eggs than nymphs and adults of *T. urticae* on rose. Regardless of prey density, *N. californicus* preferred eggs, larvae and nymphs over adult male and female *T. urticae* (Canlas *et al*., 2006). However, in the case of *O. coffeae, N. longispinosus* showed preference towards larvae and nymphs over eggs due to thickness of egg shell of *O. coffeae* (Rahman et al., 2012 a).

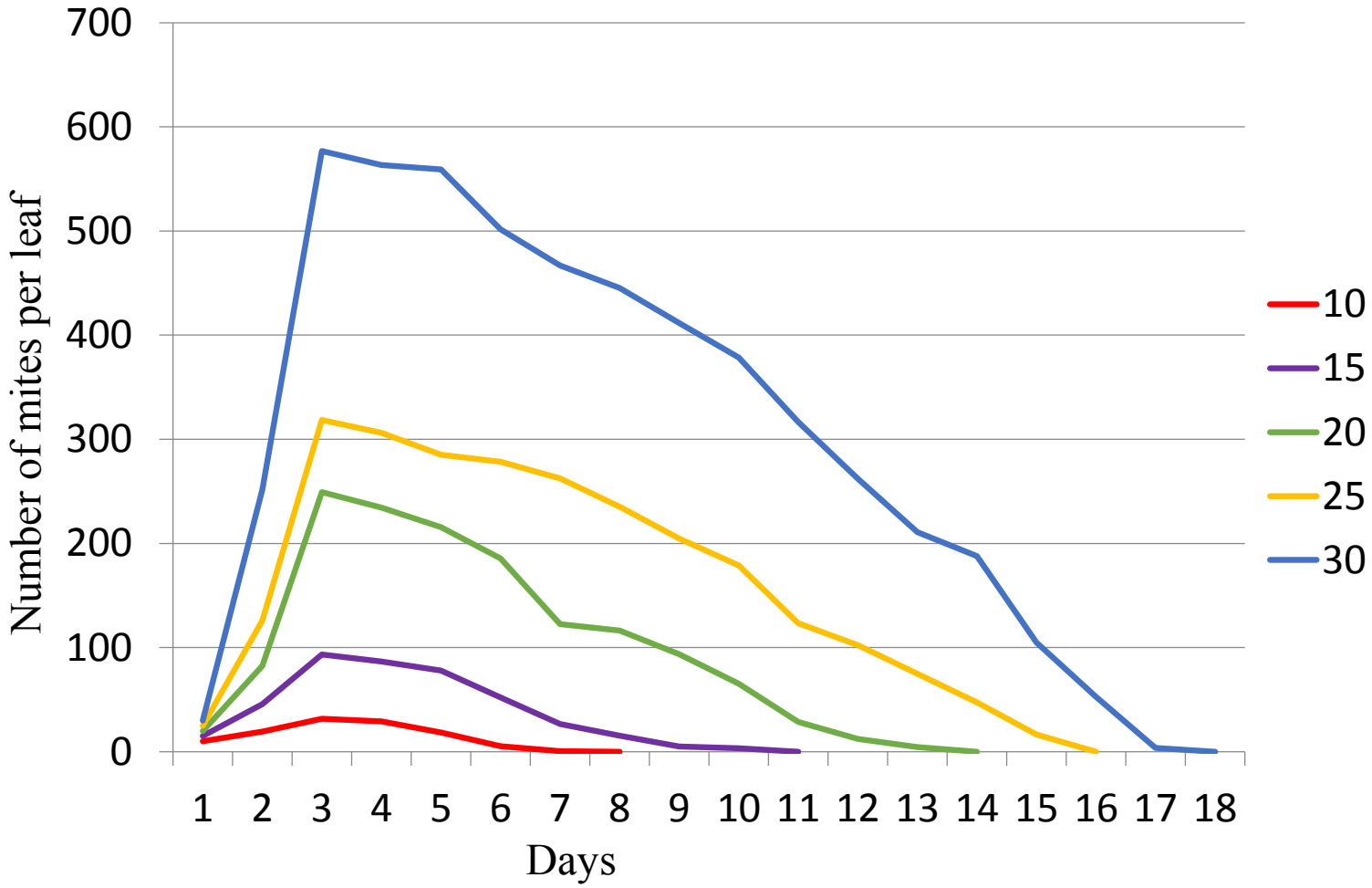


Fig. 6. Population of *Tetranychus urticae* after the release of a single predator at different prey densities

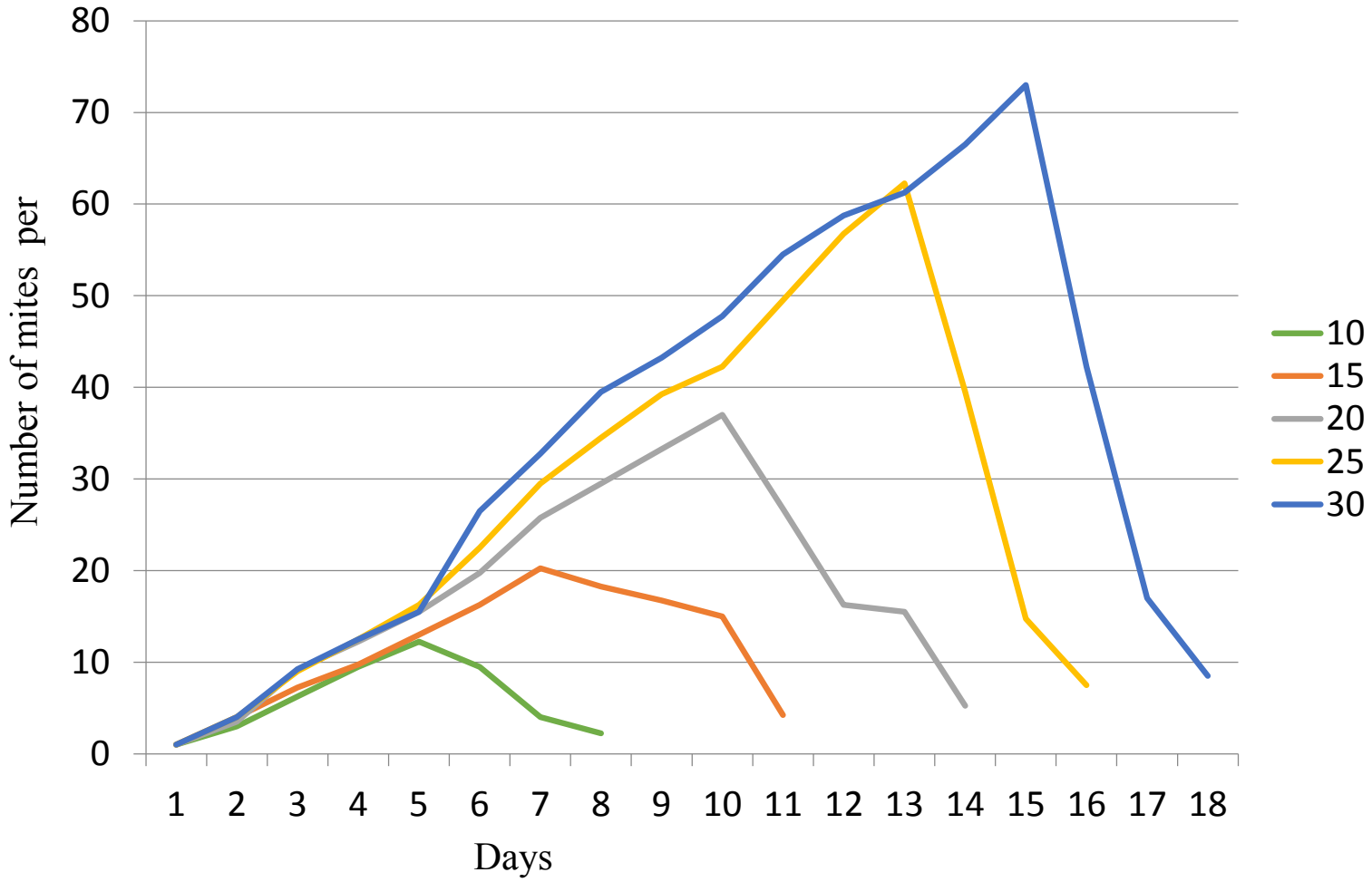


Fig. 7. Population of *Neoseiulus longispinosus* at different prey densities

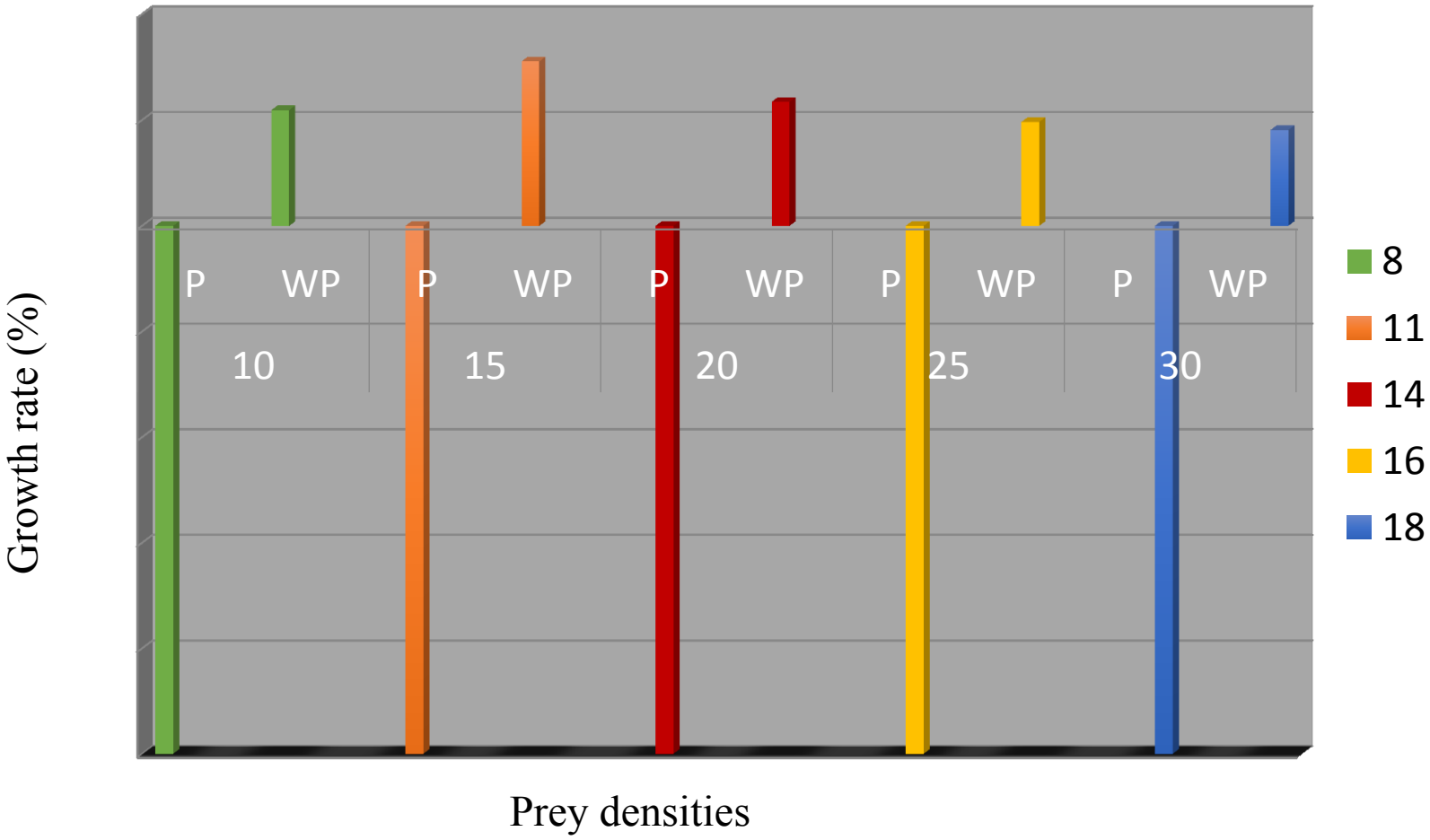


Fig. 8. Growth rate of *Tetranychus urticae* as influenced by *Neoseiulus longispinosus*

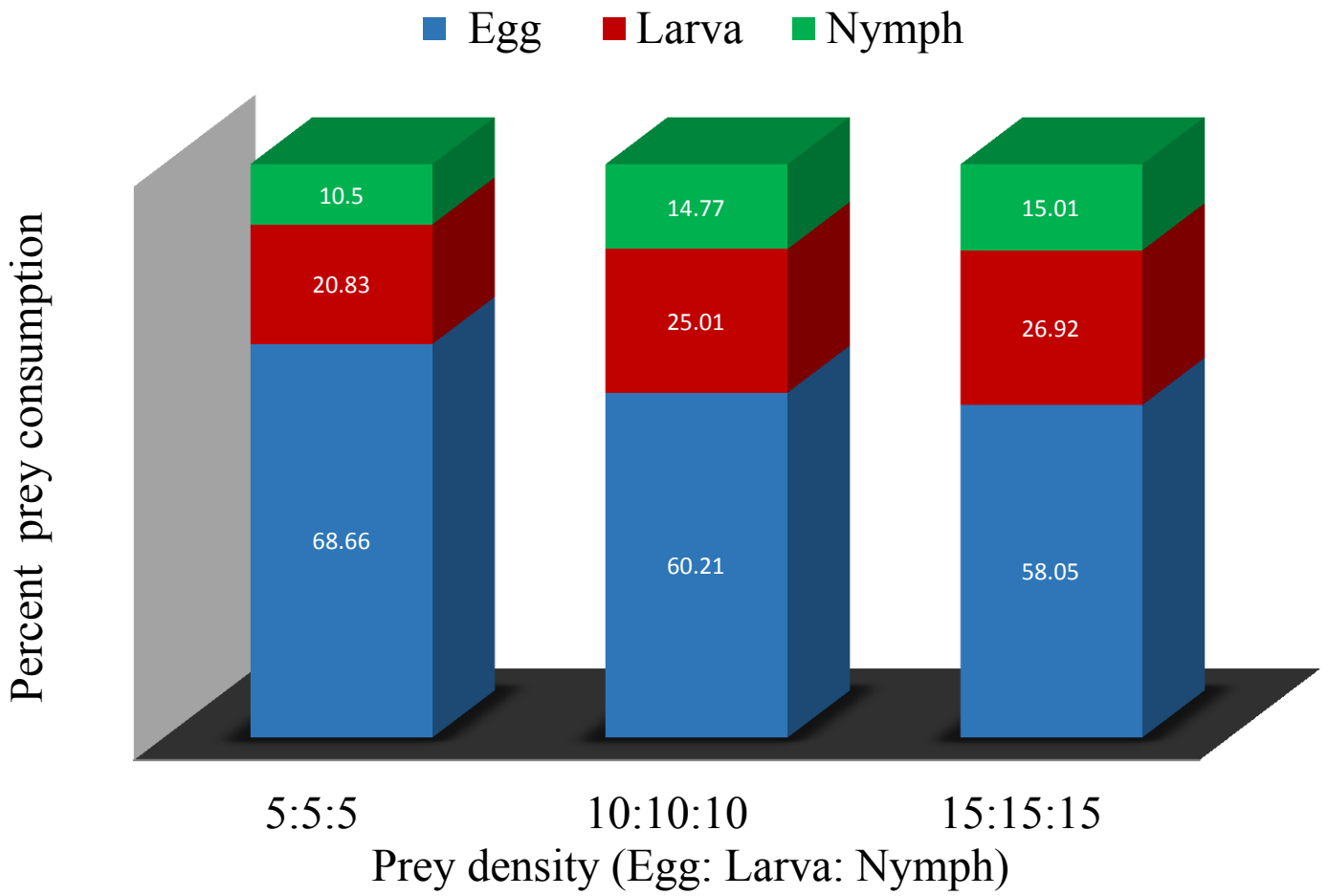


Fig. 9. Preference of *Neoseiulus longispinosus* deutonymph towards different life stages of *Tetranychus urticae*

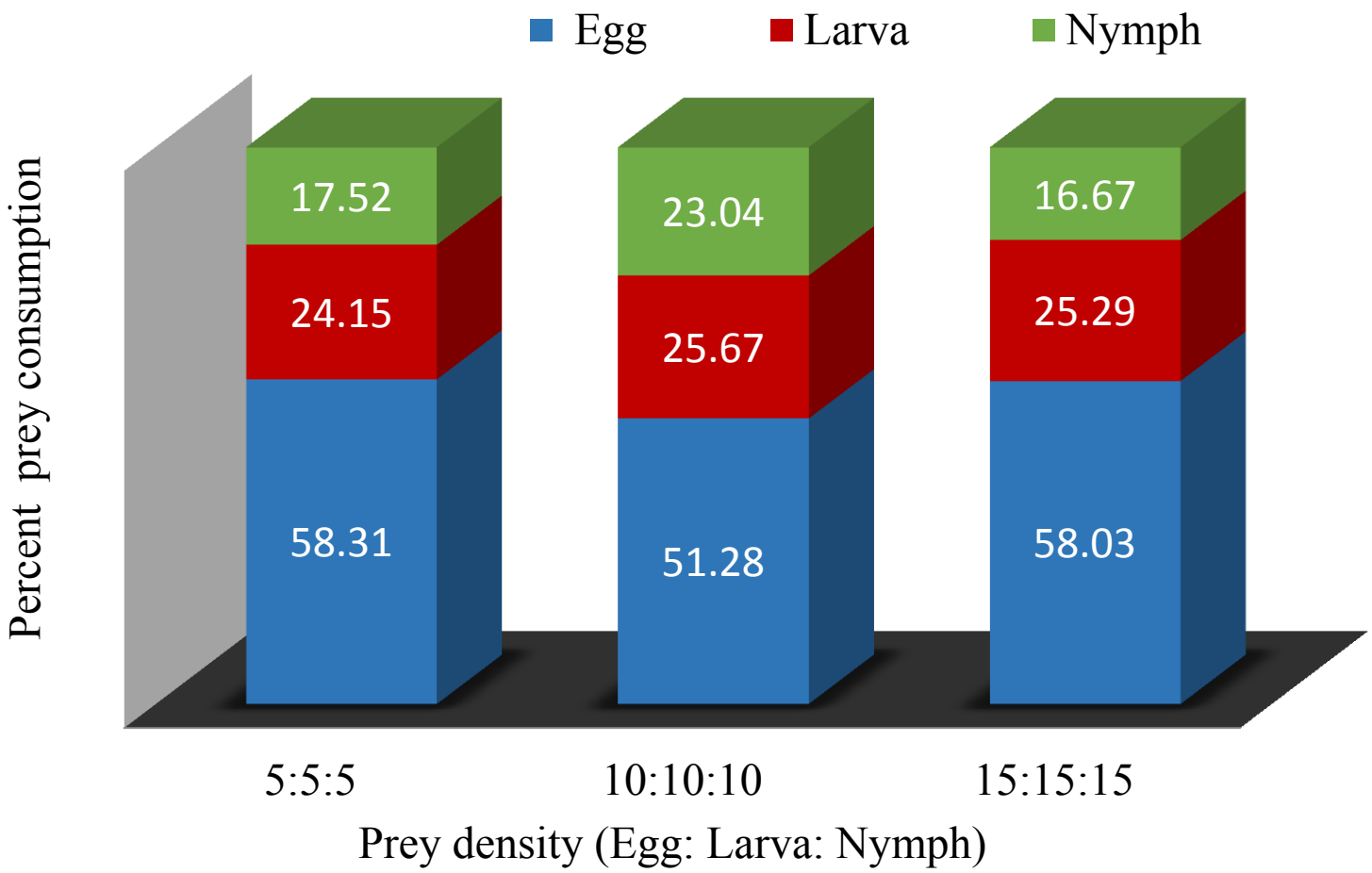


Fig. 10. Preference of *Neoseiulus longispinosus* adult towards different life stages of *Tetranychus urtic*

Jayasinghe and Mallik (2014) also indicated that predators prefer eggs than other stages of the spider mite.

Tetranychus urticae lay eggs in groups which remain close to each other. This enables the predator to spend much shorter time in search of the prey which also increases the chances of encountering the prey. As nymphs move around actively, the predator may take more time in search of the nymphal stage and this becomes the less preferred stage of the predator. Between nymph and larva, the predator preferred larva. It is found that larval stage though a mobile stage is less active compared to nymphal stage. This may be the reason for the increased preference towards larva compared to nymph. The preference shown by *N. longispinosus* towards the eggs of *T. urticae* is a desirable attribute in field, as this would help in directly hindering the proliferation of the pest.

5.5. ESTIMATION OF OPTIMUM PREDATOR: PREY RATIO

5.5.1. Laboratory Evaluation of Optimum Predator: Prey Ratio

The present study indicated that at a narrower predator: prey ratio of 1: 5 and 1: 10, the predator could eliminate the prey population in seven days and ten days, respectively. However, at these ratios, a drastic decline in the predator population was also noticed probably due to the insufficiency of food to sustain the predator population (Fig. 11). At wider ratios (1: 20 to 1: 100), total elimination of prey population could not be effected up to ten days after predator release. At a narrow predator: prey ratio, the predator density is higher than that at a wider ratio and at higher predator density it is faster elimination of prey population is only to be expected. The study also indicated that, at narrow ratios, the predator population also declined at a faster rate compared to those at wider ratios. Considerable reduction in prey population after the release of predator at narrow predator: prey ratio might have hindered the development of predator due to the non availability of prey. This might be the reason for the drastic

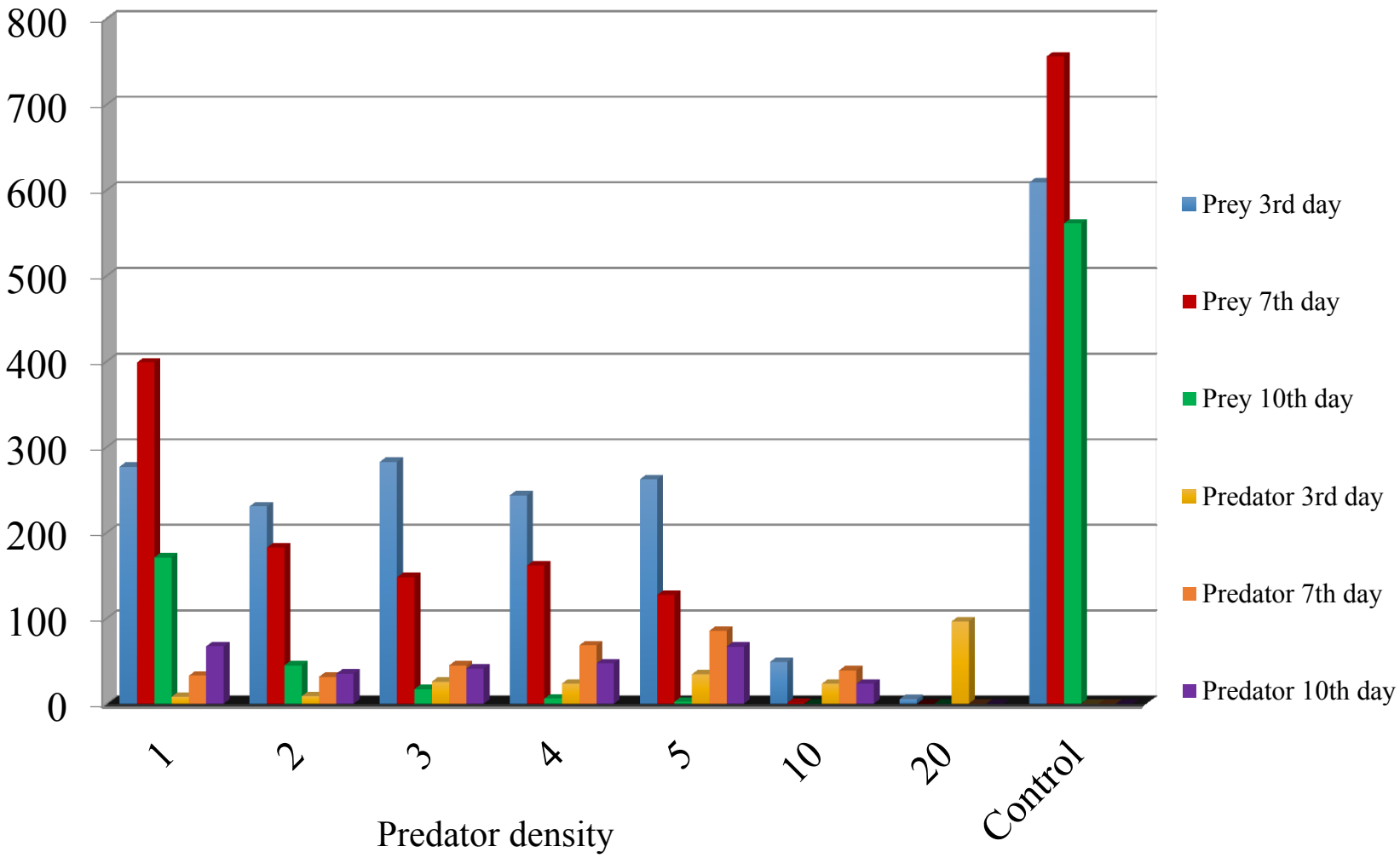


Fig. 11. Influence of predator: prey ratio on the population build up of *Tetranychus urticae* and *Neoseiulus longispinosus* on cucumber in the laboratory

reduction in predator population from seventh day onwards at the lower ratios. On the tenth day, though the prey population was not completely eliminated at the ratios 1: 20, 1: 25 and 1: 33, there was a significant reduction in the prey population. The predator population was also available in good numbers. Hence it is expected that the predators would consume the available prey mite and bring about further reduction in population. The ratios between 1: 20 and 1: 33 are found ideal as indicated by the present study. Studies by Kongchuensin *et al*. (2006) showed that the optimum predator prey ratio effective in the field was 1: 20- 1: 40. The results of Rahman *et al.* (2012 a) also indicated 1: 33 and 1: 50 as the best ratios in the laboratory for *N. longispinosus* on *O. coffeae*.

In the present study though narrow ratios were found to be effective and fast in controlling the prey mites, it is not economically viable to use predators at high densities for biocontrol programme in field.

5.5.2. Polyhouse Evaluation of Optimum Predator: Prey Ratio

In the polyhouse, the best predator: prey ratio identified were 1: 20 and 1: 25 to suppress the pest population (Fig. 12). Fifteen days after the second release of the predator, a considerable reduction in prey mite population was effected at these ratios. There was also an increase in predator population at ratios of 1: 20 and 1: 25. Further reduction in population of *T. urticae* is expected on cucumber in polyhouse by the action of these predators. Greenhouse studies conducted by Rahman *et al.* (2012 a) revealed 1: 25 as a suitable predator prey ratio to suppress *O. coffeae* on tea by *N. longispinosus*. Rajashekharappa (2010) also suggested ratios of 1: 25 or lower as the effective and economical ratio to control spider mites on rose and carnation.

The present study suggests that *N. longispinosus* can be an efficient biocontrol agent against spider mites due to several positive traits identified in the biology and predation efficiency. The predator completes its life cycle in a span as short as 4.2 days

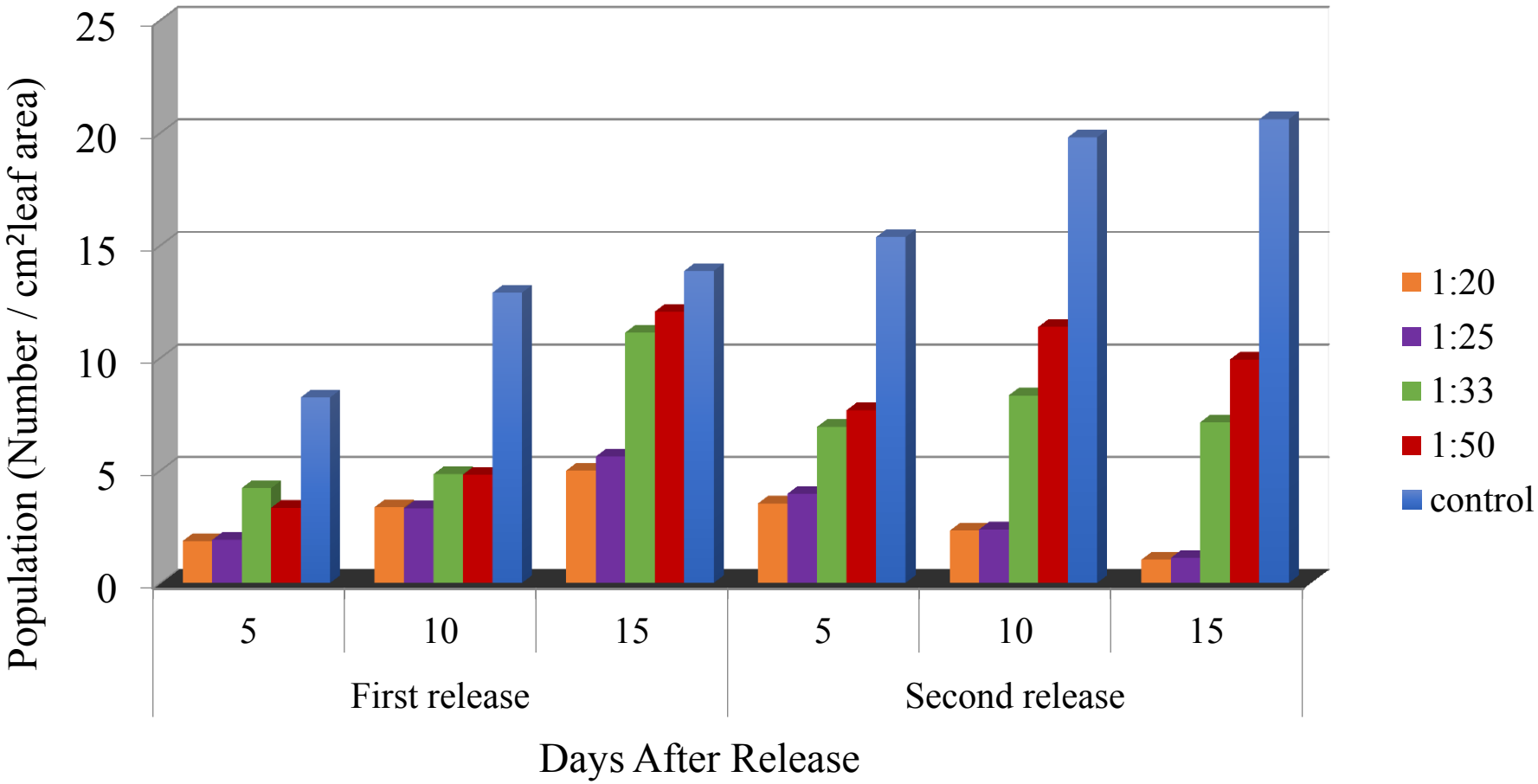


Fig. 12. Influence of predator: prey ratio on the population of *Tetranychus urticae* on cucumber in polyhouse condition

for females and 3.91 days for males compared to the longer duration of 6.75 and 7.15 days for male and female *T. urticae*, respectively. Female biased sex ratio (1: 3.31) and longer oviposition period (19.91 days) along with shorter life cycle in *N. longispinosus* would result in faster multiplication of the predator. In addition, longer life span of the adults of *N. longispinosus* (19.66 days for male and 22.75 days for female) would influence the predation efficiency as the study revealed that adults are more potent than nymphal stages. Also the preference shown by *N. longispinosus* towards the egg stage of the prey would help prevent the proliferation of the pest, there by bringing about effective management. Above all, the density dependant nature of the prey predator relationship, brought out in the present study could provide a platform for viable biocontrol strategy based on *N. longispinosus* for management of spider mites under protected cultivation in Kerala.

Summary

6. SUMMARY

The present study was undertaken at the Acarology laboratory, Department of Agricultural Entomology, College of Horticulture, Vellanikkara during 2013-2016. The thrust area of the investigation was to study the biology, incidence, crop phenology relationship and natural enemies of the two spotted spider mite *Tetranychus urticae* Koch infesting cucumber in polyhouse; biology, efficacy and prey stage preference of the predator, *Neoseiulus longispinosus* (Evans) on *T. urticae* as well as standardization of optimum predator: prey ratio of *N. longispinosus* for biological control of *T. urticae* in polyhouse.

The salient findings of the study are summarized hereunder.

- Purposive surveys conducted in fifteen polyhouses of four districts of Kerala *viz.*, Palakkad, Wayanad, Thiruvananthapuram and Thrissur revealed the occurrence of four species of tetranychid mites, *Tetranychus truncatus* Ehara, *T. urticae*, *Tetranychus okinawanus* Ehara, *Eutetranychus orientalis* (Klein) and one species of tarsonemid mite, *Polyphagotarsonemus latus* (Banks) on cucumber.

- *Tetranychus truncatus* was found to be the predominant spider mite species associated with cucumber in polyhouse and was recorded from all the districts surveyed.

- Molecular characterization was carried out for *T. truncatus* and *T. okinawanus* to confirm the species identity. On submission of DNA sequences to iBOL for barcoding, process id was received for *T. truncatus* (TOCRF001-15) and *T. okinawanus* (TOIR001-15).

- Fixed plot surveys were carried out at fortnightly intervals in selected five polyhouses of Thrissur district during June 2013 to May 2014 to study the relationship of crop stage and mite incidence on cucumber. The population of spider mites was significantly higher during the late fruiting stage of the crop, followed by early vegetative stage, late vegetative stage, flowering stage and early fruiting stage.

- A total of ten species of predators were recorded during the survey in association with spider mites of which, four were insect predators and six were mite predators. The insect predators recorded were *Oligota* sp., *Stethorus pauperculus* (Weise), *Scolothrips* sp. and a species of Cecidomyiidae. The predatory mite fauna associated with phytophagous mites on cucumber were *Neoseiulus longispinosus* (Evans), *Agistemus garrulus* Chaudhari, *Tydeus gossabaensis* Gupta, *Amblyseius paraaerialis* (Muma), *Cunaxa* sp. and *Cheyletus* sp.. *Neoseiulus longispinosus* was recorded as the dominant predatory mite associated with *T. urticae* on cucumber.

- Studies on the biology of *T. urticae* conducted at 27.05 ± 0.68ºC and 90.12 ± 6.88 per cent relative humidity recorded an incubation period of 2.85 and 2.48 days, larval period of 0.56 and 0.70 days, protonymphal period of 0.57 and 0.52 days, deutonymphal period of 0.71 and 0.77 days and total development period of 6.75 and 7.15 days, respectively, in male and female *T. urticae*.

- Adult male of *T. urticae* lived for 8.95 days while mated and unmated female lived for 11.59 days and 13.04 days, respectively. The pre- oviposition period for mated and unmated female lasted for 2.16 days and 1.78 days, respectively. Oviposition and post-oviposition periods lasted for 8.15 days and 1.67 days in case of mated female and 10.16 days and 1.86 days in case of

unmated female. Fecundity of mated female and unmated female were 47.91 and 36.08 eggs, respectively. The sex ratio was female biased (1:4.6) and the viability of eggs recorded was 92.55 per cent for mated female and 90. 23 per cent for unmated female.

- The biology studies of *N. longispinosus* showed that the life cycle consisted of five different stages such as egg, larva, protonymph, deutonymph and the adult.

- *Neoseiulus longispinosus* recorded an incubation period of 1.46 and 1.57 days, larval period of 0.60 and 0.65 days, protonymphal period of 0.86 and 0.98 days, deutonymphal period of 0.98 and 1.05 days and total development period of 3.91 and 4.27 days, respectively, in male and female.

- Adult *N. longispinosus* exhibited sexual dimorphism. The male was elongate and smaller compared to female that was dark brown in colour and bigger in size.

- Adult male of *N. longispinosus* lived for 19.66 days while female lived 22.75 days. The mean pre- oviposition, oviposition and post- oviposition period of female were 1.97, 19.91 and 0.99 days, respectively. On an average a single female laid 31.33 eggs out of which 89.83 per cent of the eggs hatched out to larva. The progeny consisted of both males and females in the ratio 1: 3.31. Parthenogenesis was not observed.

- The studies on the predatory potential of *N. longispinosus* revealed that adults consumed more prey compared to nymph at all prey densities. The predation

showed a positive correlation with prey densities with highest predation at a prey density of 40.

- The experiment conducted to study the time needed to control the prey population revealed that the predator took eight days to eliminate the prey completely at initial prey density of ten mites while at a density of thirty mites, the predator took 18 days to eliminate the prey. The result clearly indicated that the time needed to control the prey increased with increase in density of the prey.

- The results of the experiments on prey stage preference indicated that the predator, both deutonymph and adult showed significantly higher preference towards eggs at all the prey densities tested. At higher prey densities of 10:10:10 (30) and 15:15:15 (45), the predator nymph preferred larval stage of prey over the nymph.

- To standardize the optimum predator: prey ratio required for field release of *N. longispinosus* against *T. urticae* on cucumber; experiments were laid out in the laboratory and polyhouse. In the laboratory, narrow prey ratios of 1: 5 and 1: 10 were found to be superior over wider ratios in eliminating the prey mite at a faster rate. In the polyhouse, the predator: prey ratios of 1: 20 and 1: 25 were identified as the optimum ratios to bring about significant reduction in spider mite population on cucumber.

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Appendix

Appendix. 1 Temperature and humidity recorded during the study period

Date	Temperature (º C)	Humidity (%)
Biology of *Tetranychus urticae*		
06/06/13	28.0	80
07/06/13	26.9	87
08/06/13	26.9	92
09/06/13	26.9	92
10/06/13	26.9	92
11/06/13	25.5	98
12/06/13	26.2	97
13/06/13	25.5	90
14/06/13	26.4	99
15/06/13	26.9	97
16/06/13	26.3	96
17/06/13	27.4	94
18/06/13	27.4	92
19/06/13	26.5	97
20/06/13	26.8	94
21/06/13	27.3	92
22/06/13	26.2	96
23/06/13	28.0	75
24/06/13	27.6	76
25/06/13	27.6	85
26/06/13	28.8	77
27/06/13	27.5	85
28/06/13	27.5	84
29/06/13	26.9	89
30/06/13	27.1	87
01/07/13	26.4	93
02/07/13	26.1	99
03/07/13	26.6	97
04/07/13	27.4	97
05/07/13	26.7	97
06/07/13	26.8	94
07/07/13	27.5	89
08/07/13	27.0	91
09/07/13	27.2	95
10/07/13	27.6	89
11/07/13	27.8	93
12/07/13	26.9	90

13/07/13	27.9	77
14/07/13	27.2	80
15/07/13	27.9	81
Mean	27.05	90.125
Biology of *Neoseiulus longispinosus*		
28/12/13	29.2	73
29/12/13	29.2	71
30/12/13	28.5	77
31/12/13	28.2	80
01/01/14	28.5	79
02/01/14	28.9	83
03/01/14	27.7	83
04/01/14	27.9	79
05/01/14	27.5	87
06/01/14	26.2	87
07/01/14	26.8	88
08/01/14	26.5	94
09/01/14	27.00	89
10/01/14	27.4	86
11/01/14	27.2	59
12/01/14	27.4	75
13/01/14	29.2	70
14/01/14	29.1	81
15/01/14	28.9	80
16/01/14	29.00	69
17/01/14	29.50	71
18/01/14	29.20	82
19/01/14	27.50	87
20/01/14	28.50	80
21/01/14	27.90	86
22/01/14	27.40	83
23/01/14	28.40	87
24/01/14	28.80	90
25/01/14	27.50	78
26/01/14	27.90	82
Mean	28.05	79.68

EFFICACY OF *Neoseiulus longispinosus* (EVANS) (MESOSTIGMATA: PHYTOSEIIDAE) FOR THE MANAGEMENT OF *Tetranychus urticae* KOCH (PROSTIGMATA: TETRANYCHIDAE) ON CUCUMBER UNDER PROTECTED CULTIVATION

by

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ABSTRACT OF THE THESIS

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ABSTARCT

A study was undertaken at the Acarology laboratory, Department of Agricultural Entomology, College of Horticulture, Vellanikkara during 2013-2016, to investigate the biology, incidence, crop phenology relationship and natural enemies of the two spotted spider mite, *Tetranychus urticae* Koch infesting cucumber in polyhouse as well as to study the biology, efficacy and prey stage preference of the predator, *Neoseiulus longispinosus* (Evans) on *T. urticae* and standardize the optimum predator: prey ratio of *N. longispinosus* for biological control of *T. urticae* in polyhouse.

Purposive surveys, conducted in fifteen polyhouses in four districts of Kerala *viz.*, Thrissur, Palakkad, Wayanad and Thiruvananthapuram, revealed the occurrence of four species of tetranychid mites, namely, *Tetranychus truncatus* Ehara, *T. urticae* Koch, *T. okinawanus* Ehara, *Eutetranychus orientalis* (Klein) and one species of tarsonemid mite, *Polyphagotarsonemus latus* (Banks) on cucumber. The occurrence of *T. truncatus* and *T. okinawanus* is a new report on cucumber in polyhouse. Hence, DNA barcoding was carried out to confirm the species identity of *T. truncatus* (TOCRF001-15) and *T. okinawanus* (TOIR001-15). Studies on the relationship of crop stage and mite incidence on cucumber revealed that the population of spider mites was significantly higher during the late fruiting stage of the crop, followed by early vegetative stage. Relatively lower population was recorded at flowering stage and early fruiting stage.

Four species of insect predators and six species of mite predators were recorded in association with spider mites on cucumber. The insect predators were *Stethorus pauperculus* (Weise), *Oligota* sp., *Scolothrips* sp. and an unidentified species of Cecidomyiidae. The predatory mite fauna included *Agistemus garrulus* Chaudhari, *Amblyseius paraaerialis* (Muma), *Cunaxa* sp., *Cheyletus* sp., *Neoseiulus longispinosus* (Evans) and *Tydeus gossabaensis* Gupta. *Neoseiulus longispinosus* was

found to be the predominant species of predatory mite on spider mites infesting cucumber.

Tetranychus urticae recorded a developmental period of 6.75 days in male and 7.15 days in female. Adult male lived for 8.95 days while mated and unmated female lived for 11.59 days and 13.04 days, respectively. Mated and unmated females recorded a fecundity of 47.91 and 36.08 eggs, respectively. The sex ratio was female biased (1:4.6) in *T. urticae*.

Total developmental period of *N. longispinosus* was 3.91 and 4.27 days for male and female, respectively. Adult male lived for 19.66 days and the female, for 22.75 days. On an average, a single female laid 31.33 eggs and the progeny consisted of both males and females in the ratio 1:3.31. Parthenogenesis was not observed in *N. longispinosus*.

The adult of *N. longispinosus* recorded significantly higher predation compared to the nymph. Both nymph and adult, showed preference towards egg compared to active stages of the prey. The time needed to eliminate the available prey population was found to increase with increase in prey density.

Studies conducted to identify the optimum predator: prey ratio required for field release of *N. longispinosus* against *T. urticae* on cucumber in the laboratory showed that, at ratios of 1:5 and 1:10, the prey population was completely eliminated by tenth day. The prey population recorded in the ratios, 1:20, 1:25 and 1:33 were on par with this. In the polyhouse, the predator: prey ratios of 1:20 and 1:25 were found to significantly reduce the population of *T. urticae* on cucumber.

The present study has revealed the potential of the predatory mite, *N. longispinosus* as a biocontrol agent of the spider mites. The short life cycle, longer life span of adults, female biased sex ratio and preference for egg stages and above all, the density dependant nature of the prey predator relationship, brought out in the present study could provide a platform for viable biocontrol strategy based on *N. longispinosus* for management of spider mites under protected cultivation in Kerala.