Eco-Toxicological Studies on the Solenopsis Mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Coccoidea: Pseudococcidae) and its Predators

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Eco-Toxicological Studies on the Solenopsis Mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Coccoidea: Pseudococcidae) and its Predators

A Thesis

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This is to certify that the thesis entitled “Eco-Toxicological Studies on the Solanum Mealybug, Phenacoccus solenopsis Tinsley (Hemiptera: Coccoidea: Pseudococcidae) and its Predators” submitted to the Post-Graduate School, Indian Agricultural Research Institute, New Delhi, in partial fulfilment of the requirements for the degree of Doctor of Philosophy in Entomology, embodies the results of bonafide research work carried out by Mr. Sachin Suresh Suroshe under my guidance and supervision. No part of the thesis has been submitted for any other degree or diploma.

All the assistance and help received during the course of the investigation have been duly acknowledged by him.

(R. D. Gautam)
Chairman
Advisory Committee
Dedicated to my
grand-mother
late smt. Rukhmabai Chavan,
father
late shri. Suresh Suroshe
and mother
smt. Pushpa Suroshe
Imagination is more important than knowledge:

Albert Einstein
1. INTRODUCTION

Pseudococcidae or Mealybugs occur in all zoogeographic regions of the world and are abundant in most ecosystems. There are about 2000 species of mealybug occurring on about 250 plants families throughout the world (Ben-Dov et al., 2011). In India, more than 100 species of pseudococcids are known to attack a variety of plant species (Varshney, 1985). Interestingly, the present solenopsis mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Coccoidea: Pseudococcidae), which is an emerging pest problem in Indian subcontinent does not figure in the list given by Varshney in 1985.

*P. solenopsis*, a pest of ornaments and fruit trees worldwide and to be cryptic in nature. It was described originally from the U.S. in 1898 and remained widespread only in the U.S. until 1992 (Ben-Dov, 2004). In 1992, it was reported in Central America, the Caribbean and Ecuador (Williams & Granada de Willink, 1992). Larraín (2002) noted the species, *P. solenopsis* as a pest of sweet pepinos, *Solanum muricatum* Aiton in Chile for the first time. Mark and Gullan (2005) reported it from the state of Espirito Santo, Brazil, where it was found infesting tomato for the first time. Recently, it is reported as first record on *Hibiscus rosa-sinensis* L. in Nigeria (Akintola and Ande, 2008). It is uncertain if this sequence represents a true, recent expansion in geographic distribution of the species from North America to South America or is simply a coincidence of collection and identification efforts as well as spread potential of *Phenacoccus* spp (Gautam, 2007). The species, *Phenacoccus* has been found previously on a relatively wide variety of host plants including species in economically important families such as Cucurbitaceae, Fabaceae as well as Solanaceae.

Recently in India, mealybug, *P. solenopsis* and *Phenacoccus solani* Ferris (Hemiptera: Pseudococcidae) were observed on several plant hosts including cotton (Jhala, et al. 2008). Solanum mealybug, *P. solani* (Pseudococcidae: Homoptera) was reported attacking 64 plant species belonging to 27 families, including weeds in and around Delhi (Gautam, 2007). To combat the pest menace, pesticides of worth over Rs. 500 crores were sold in Punjab (India) in just three months from June to August 2007 (Dutt, 2007). *P. solenopsis* is very similar in appearance (microscopically) to *P. solani*
and *Phenacoccus defectus* Ferris (Williams & Granada de Willink 1992). However, the live appearance of *P. solenopsis* differs from these other two species in that the adult female of *P. solenopsis* generally has paired dark spots and/or stripes dorsally, whereas the females of the other two species appear to be uniformly white dorsally (Miller et al. 2005). *P. solenopsis* usually has short lateral wax filaments and slightly longer terminal wax filaments (less than half as long as the body).

The detailed studies on seasonal morphological variation of mealybug attacking cotton and other crops in the Indian sub-continent and elsewhere in Asia showed it to be a *P. solenopsis*. It was concluded that this species shows considerable morphological variation, particularly in the number of the multilocular disc pores on the submargin of the abdomen and in the frequency of the oral collar tubular ducts throughout the body and that this variation appears to be environmentally induced (Hodgson et al. 2008). Reproductive potential of *Maconellicoccus hirsutus* (Green) was more hence it out numbered other two spp. namely *P. solenopsis* and *P. solani* by 4th generation in the laboratory (Sudhida et al. 2009). Hence, identity crisis of *Phenacoccus gossypiphilous* (Stanley), *P. solenopsis* and *P. solani* remains a matter of controversy in the Indian subcontinent.

As this mealybug is thought to be an exotic one and might have got introduced recently, the knowledge about its biology, natural enemy complex and management tactics is scanty in Indian conditions. Against this backdrop, in order to throw light on above said aspects thesis entitled “Eco-Toxicological Studies on the Solenopsis Mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Coccoidea: Pseudococcidae) and its Predators” was taken up in the Biological Control Laboratory, Division of Entomology, Indian Agricultural Research Institute, New Delhi, during 2006-09, under the following objectives:

1. To study the natural enemy complex of *P. solenopsis* infesting different plant hosts.
2. To study the population dynamics of *P. solenopsis* and its natural enemies associated with the brinjal, *Solanum melongena*; gurhal, *Hibiscus rosasinensis* and sorrel, *Hibiscus sabdarifa*. 
3. To study the biology of *P. solenopsis* on different hosts in the laboratory.
4. To elucidate the effect of recommended insecticides on the important predators and its prey *P. solenopsis*.
5. To elucidate the effect of recommended insecticides on the solitary endoparasitoid (*Aenasius bambawalei* and its hyper-parasitoid, *Marietta leopardina*) of *P. solenopsis*.

1.1 Background

1.1.1 Mealybugs in general

Mealybugs (Hemiptera: Pseudococcidae) are cottony in appearance, small oval, soft-bodied sucking insects. Adults are found on leaves, stems and roots and are covered with white mealy wax, which makes it difficult to eradicate. They form colonies on stem and leaves developing into dense, waxy, white masses. They suck a large amount of sap from leaves and stem with the help of piercing and sucking mouth parts, depriving plants of essential nutrients. A byproduct of mealybug feeding is sticky honeydew which coats infested foliage, and provides an excellent medium for growth of the black sooty mold fungi. This black coating further renders affected plants unsightly (Osborne, 2011).

Mealybugs have 4 female instars and 5 instars in the male. Eggs or first instars are laid by the adult female. Eggs are normally laid in an ovisac that can enclose all or part of the body of the female. Most species that lay first instars rather than eggs lack any substantial ovisac. Even though the majority of species have legs in all instars, most mealybugs remain relatively stationary throughout their life; a few species such as some members of the genus *Phenacoccus*, move to different areas of the host for overwintering, feeding, oviposition, and molting. Most species have 1 or 2 generations a year, although some are reported to have as many as 8 generations in the greenhouse. Both parthenogenetic and sexual species are common (Ben-Dov *et al.*, 2011).

1.1.2 Worldwide first reports of *Phenacoccus spp.*

Fuchs *et al.*, (1991) reported for the first time *P. solenopsis* infesting cotton in USA. During surveys in Texas, the mealybug was found to infest 29 other plant species
in 13 families. Mazzeo et al., (1999) reported *P. solani* as a new record in Italy (Sicily) on *Encephalartos* sp. (Cycadaceae) in a cold greenhouse. Larrain (2002) noted *P. solenopsis*, for the first time as a pest of *Solanum muricatum* in IV region, Chile, where it is known as soil mealybug. Granada de Willink and Maria (2003) reported *P. solenopsis* for the first time from Argentina. The mealybug, *P. solani*, endangering many hosts was found in Taiwan for the first time (Chen et al., 2002). Jansen (2004) recorded *P. solenopsis* for the first time in 1993 on solanum from non commercial green houses in the Netherlands. Specimens collected by Moghaddam et al., (2004) were identified as *P. solani* infesting *Festuca* sp. and *Chrysanthemum morifolium* in Iran for the first time. Pellizzari and Russo (2004) along with a list of the scale insects recorded *P. Solani* the exotic introduced species from Sicily, Italy.

Ben Dov (2005) recorded solanum mealybug, *P. solani* from Israel for the first time on plants belonging to family Amaryllidaceae, Compositae and Solanaceae. Prishanthini and Laxmi Vinobaba (2009) identified *P. solenopsis* for the first time in Sri Lanka at the Department of Zoology of the Eastern University, it was observed on home garden plants in June 2008.

### 1.1.3 Recent outbreaks of *P. solenopsis* in India

*P. solenopsis* has been reported from 35 localities of various ecological zones of the globe (Ben Dov et al., 2009). *P. solenopsis* has been the current topic of research for insect taxonomists and applied entomologists in India due to its invasiveness, rapid spread, morphological and biological variations and the need for establishing an effective control strategy (Vennila et al., 2010a). Survey across 47 locations of the country between months of late 2007 and early 2008 established the predominance of *P. solenopsis* in India (Nagrare et al., 2009). In the current decade, increased build up of various mealybug species in crop plants and wild plants is observed mainly due to certain abiotic changes in climate and environment. During the last few years, mealybugs which were considered to be minor pests in many crops have acquired the status of major pests especially in cotton, vegetables and fruits. *P. solenopsis* has recently emerged as a serious insect pest in India, attacking important field crops such as cotton, okra, tomato, brinjal, chilli and ornamental plants such as *Hibiscus rosa-sinensis,*
particularly in the cotton growing belts of Punjab, Haryana, Gujarat, Maharashtra and Andhra Pradesh (Dhawan et al., 2007; Tanwar et al., 2007; Jhala et al., 2008 and Bhosle et al., 2009).

1.1.3 Economic impact of *P. solenopsis* on Indian Agriculture

Pesticides worth over Rs. 500 crores were sold in Punjab during cotton growing season of 2007, in order to check infestation of this mealybug. The cost of plant protection to farmers on account of mealybug attack on B.T. cotton in Punjab has increased by Rs 2500 per acre (Gautam, 2008a). During 2007, *P. solenopsis* spread to major cotton growing belts of Punjab and caused 30-40 per cent yield loss in cotton (Dhawan et al., 2007). There was an outbreak of *P. solenopsis* on cotton during October-December, 2006 in middle Gujarat (Jhala et al., 2008). A roving survey carried out in major cotton growing districts of Marathwada region of Maharashtra during crop season 2007-08 revealed that the severity of *P. solenopsis* infested bolls ranged between 10.10 to 35.37 per cent (Bhosle et al., 2009). Infestation of *P. solenopsis* at most of the places in North and Central zones ranged from mild (10-20%) to high (40-60%) during 2007 and 2008 (Tanwar et al., 2011).

The detail review of previous work done in line with the present study was made and presented under the following headings and subheadings.

1.2 Research area I:

Natural enemy complex of *P. solenopsis*

1.2.1 Natural enemies of *Phenacoccus* spp.

Pachyneuron sp., Prochiloneurus aegyptiacus and Prochiloneurus annulatus). A survey of parasitoids in Malda (West Bengal) on mango revealed Chartocerus sp., Azotus sp. and Gyranusoidea tebygi on Rastrococcus invadens. Chartocerus walkeri, Aprostocetus sp., Promuscidea unfasciiventris, Anagyrus pseudococci and Anagyrus mirzai were recorded on Rastrococcus iceryoides. Two species of Aprostocetus, Encyrtus sp. and Austroterobia maldica were recorded on Icerya minor. Coccophagus ceroplastae and P. unfasciiventris were recorded from Pulvinaria polygonata (Das and Sahoo, 2005).

Hayat et al., (2007) reported 7 spp. of chalcids (Myiocnema comperei, P. unfasciiventris, Prochiloneurus pulchellus, Neocaritopus orientalis, Leptomastix gunturiensis and Leptomastix nigrocincta) belonging to Aphelinidae, Encyrtidae and Eulophidae on mealybug, Coccidohystrix sp. infesting Abutilon indicum, in Aligarh, Uttar Pradesh. The genus Myiocnema is a new record for India. Gautam et al., (2007) reported five predatory coccinellids viz; Brumoides lineatus, Brumoides suturalis, Cheilomenes sexmaculata, Nephus regularis and Scymnus coccivora from the field population of P. solani infesting different plant species in Delhi. The unidentified encyrtid and dipteran parasitoids were also present. Encyrtid parasitoids reared by Daane et al., (2008) from either obscure mealybug, Pseudococcus viburni (Signoret), or Planococcus longispinus (Targioni Tozzetti) were A. pseudococci (Girault), Leptomastix dactylopii Howard, L. abnormis (Girault), Coccidoxenoides perminutus Girault, and Tetracnemoidea peregrina (Compere). A hyperparasitoid, Chaetocerus sp. was also reared during surveys on grapes in California (USA). Survey, in Delhi during July and August, 2008 indicated the presence of cocoons of a hymenopterous parasitoid, Aenasius sp. (Chalcidoidea: Encyrtidae) on mealybug, P. solenopsis. Natural parasitism was observed from 20-70 per cent. This parasitoid was a solitary endoparasitoid causing mealybugs to swell and change their color to light brown (Tanwar et al., 2008).

Gautam et al., (2009) reported the phenomenon of fortuitous biological control which was observed due to presence of an encyrtid parasitoid (Aenasius sp) on exotic P. solenopsis, which parasitized 70-80 per cent mealybugs in the field while it was not recovered from locally existing mealybugs viz; Ferrisia virgata (Cockerell), M. (Phenacoccus) hirsutus (Green), Nipaecoccus viridis Newstead, Rastrococcus iceryoides Green and Planococcus citri (Risso). Hayat (2009) described a new species of
parasitoid, *Aenasius bambawalei* Hayat which was reared from *P. solenopsis*, a mealybug that has spread to all the cotton growing areas in India. Saroja *et al.*, (2009) reported natural parasitism in the range of 8-26 % of *P. solenopsis*, by two hymenopteran parasitoids *viz.*, *Aenasius* spp (Encyrtidae) and another one as unidentified species. Monthly surveys, in Haryana state on cotton infested by *P. solenopsis* recovered primary parasitoid, *A. bambawalei*, and four hyper parasitoids; the most abundant hyper parasitoid was *M. comperei* Ashmead. Out of six predators recovered, *B. suturalis* and *N. regularis* were abundant (Ram Pala and Saini, 2010). *A. bambawalei*, *A. kamali*, *Cryptolaemus montrouzieri*, *Chrysoperla carnea*, *Verticillium lecanii* and *Beauveria bassiana* are the effective biological control agents in managing the infestation of *P. solenopsis* (Joshi *et al.*, 2010). *P. solenopsis* is known to be an invasive species which was first observed in 2005 in Pakistan. The population buildup of this pest reached the highest level in 2007. The possibility that the parasitoid *A. bambawalei* in this region came with *P. solenopsis* cannot be ruled out (Muhammad Ashfaq *et al.*, 2010). Three species of parasitoids *viz.*, *A. bambawalei*, *P. unfasciaventris* Girault (Aphelinidae) and *Aprostocetus bangaloricus* Narendran (Eulophidae) were recorded on *P. solenopsis*. Five encyrtids *viz.*, *Encyrtus aurantii* (Geoffroy), *P. pulchellus* Silvestri, *Anagyrus dactylopii* (Howard), *A. mirzai* Agarwal and Alam and *Homalotylus albiclavatus* (Agarwal) and one each of Aphelinidae (*P. unfasciaventris* Girault), Signiphoridae (*Chartocerus kerrichi* Agarwal), Pteromalidae (*Pachyneuron leucopiscida* Mani) and Eulophidae (*A. bangaloricus*) were also documented as parasitoids of *M. hirsutus*. Coccinellids *viz.*, *B. suturalis*, *C. sexmaculata* (F.) *Scymnus castaneus* Sicaid and *C. montrouzreri* were found as predators of *P. solenopsis* (Vennilla *et al.*, 2010). *A. bambawalei*, which was reported as a solitary endoparasitoid of *P. solenopsis* in India, mostly preferred the third instar of mealybug for development (Fand *et al.*, 2011).

**1.3 Research area II:**

**Population dynamics of *P. solenopsis* and its natural enemies**

**1.3.1 Population dynamics of *Phenacoccus* spp.**
No correlation was detected by Ru et al., (1991) between parameters describing rainfall and variation in *Phenacoccus manihoti* Matile-Ferrero, density in Congo on cassava. Jeyakumar et al., (2009) through intensive surveillance in cotton growing belts of Punjab found that the temperature was negatively and relative humidity was positively correlated with both the incidence and the intensity of *P. solenopsis* on the cotton. Sporadic infestation of *P. solenopsis* was found during second fortnight of August in the Warangal district of Andhra Pradesh, but there was severe infestation during second week of October that persisted till the end of November in 2008 (Saroja et al., 2009). The incidence of *P. solenopsis* was studied by Dhawan et al., (2009), on cotton in Punjab; the highest field infestation recorded was mostly in the 30th meteorological week with 14.9, 31.5 and 26.9 per cent in Bathinda, Muktsar and Ferozepur districts, respectively. In Faridkot, the highest field infestation of 10.2 per cent was recorded in 34th meteorological week. Positive correlation was noticed among the per cent field infestation, number of infested rows and temperature, whereas negative correlation was observed with relative humidity and rainfall, all the meteorological parameters influenced the incidence of mealybug. Suresh et al., (2010) observed *P. solenopsis*, population was highest during June and decreased slowly during September and there was no incidence up to February. There was a significant positive correlation with minimum temperature and significant negative correlation with relative humidity. Hanchinal et al., (2010) *P. solenopsis* population was significantly and positively correlated with maximum temperature (0.775) and negatively correlated with other parameters in Bt cotton.

Potential distribution of invasive mealybug, *P. solenopsis* was analyzed using the CLIMEX model which revealed mealybug is limited by cold in high latitudes and altitudes, and dryness in northern Africa, inland Australia and parts of the Middle East. The key limiting factors are low precipitation as well as minimum temperatures in northern areas of America (Wang et al., 2010). Vennila et al., (2010b) reported that, preliminary analysis based on single year’s data indicated significant influence of rainfall in reducing the severity of *P. solenopsis* but not the incidence.
1.3.2 Population dynamics of N.E’s

Hanchinal et al., (2010) reported maximum population of coccinellids (0.14/plant) and Chrysoperla (0.13/plant) on cotton mealybug *P. solenopsis*, among the predators, Chrysopa significantly correlated with relative humidity and others were non significant. Highest parasitoids (20.65 %) were recorded during 7th meteorological week which coincides with the higher population of *P. solenopsis*; the parasitoid cocoons were positively correlated with maximum temperature but negatively correlated with other meteorological parameters. Ram Pala and Saini (2010) showed that in the absence of A. bambawalei, incidence of *P. solenopsis* was quite high on cotton in 2008. However, during 2009, as the activity of the primary parasitoid increased, the mealybug population reduced significantly by August. During mid-season (July-August), A. bambawalei was attacked by hyper parasitoids, particularly *M. comperei*, which caused considerable reduction in its population during August.

1.4 Research area III:

Biology of *P. solenopsis* on different hosts in the laboratory

1.4.1 Life history and biology of *Phenacoccus* spp.

Sinacori (1995) reported that, *Phenacoccus madeirensis* Green accomplished 5-6 generations per year and overwinters as both 1st and 2nd instar larvae. The duration of the different developmental stages at 30 + 2°C was observed as follows: egg, 1-4 days; 1st instar, 3-4 days; 2nd instar, 7-8 days; 3rd instar, 6-7 days; adult female, 5-8 days. While studying the temperature effects on Madeira mealybug, *P. madeirensis*, Juang Horng Chong et al., (2003) reported that the total developmental duration of female mealybugs was 30 days at 25 °C and of males was 3 to 9 days more than females. Female mealybugs made up 50 per cent of the adult populations and adult longevity at 25 °C was 3 and 20 days for males and ovipositing females, respectively. The females of *Phenacoccus bengalensis* started oviposition at the age of 31-42 days. The pre-oviposition and oviposition period, fecundity and incubation period of eggs were 14.20 days, 9.08 days, 67.42 eggs per female and 4.57 days, respectively. The longevity of adult female ranged from 47 to 55 days (Mishra et al., 2004). Nakahira and Arakawa
(2006) reported that, the total developmental period of immatures and pre-reproductive period of adults of *P. solani* decreased significantly with increased temperatures.

*P. solani*, completed its life cycle in 30-35 days. Mean reproductive potential (crawlers per female) on sunflower was (313.6) > hibiscus (284.2) > okra (250.8) > cotton (232) > lantana (219.6) > tomato (216.4) > chilli (198.2) > *Parthenium* (191.0) > wild jute (170.4) > *Amaranthus* (159.2) > carpet grass (154.2) > *Chenopodium* sp. (125.6) (Gautam *et al.*, 2007). Akintola and Ande (2008) conducted a survey of mealybugs, in Southern Guinea Savanna of Nigeria, during which they reported three nymphal instars (each with duration of 6, 8 and 10 days respectively) and fourth instar as a fully formed adult with total life cycle completed in 37 days. Dhawan and Saini (2009) reported that *P. solenopsis* reproduced viviparously and reached the adult stage after passing through three nymphal instars. After second instar, the males formed a cocoon of cottony white wax from which winged flying adult emerged after a period of 7-8 days. The life cycle was completed within 16-23 and 27-38 days by male and female, respectively. The average fecundity varied between 270-340 nymphs/female. Fand *et al.*, (2010a) reported the life cycle of *P. solenopsis* as 27.25 ± 0.5 days in which female produced 351 ± 44.73 young ones in her ovipositional period of 11.75 ± 0.96 days. Length of first instar nymphs of *P. solenopsis* was seen as 0.41 ± 0.02, whereas width as 0.20 ± 0.01 mm. Length and width of second instar nymphs seen as 0.79 ± 0.08 and 0.35 ± 0.04 mm, respectively. Length and width of third instar female observed as 2.05 ± 0.02 and 1.11 ± 0.06 mm, respectively. Body length of female was recorded as 4.70 ± 0.47 and width as 2.55 ± 0.14 mm. Longevity of female lasted for 33.67 ± 1.19 days while total life cycle lasted for 58.00 ± 3.72 days. Preoviposition, oviposition, and post-oviposition varied from 8.56 ± 0.61, 16.73 ± 0.57 and 9.33 ± 0.47 days, respectively (Nikam *et al.*, 2010a). Vennila *et al.*, (2010a) studied the biology of *P. solenopsis* on cotton under laboratory conditions. The developmental period from immature crawler to adult stage was greater for males (18.7 ± 0.9 days) as compared to females (13.2 ± 1.8 days). Females produced crawlers ranging from 128 and 812, with a mean of 344 ± 82. The reproductive period lasted 30.2 ± 8.2 days. Parthenogenesis with ovoviviparity (96.5%) was dominant over the oviparous (3.5%) mode of reproduction. Adult females lived 42.4 ± 5.7 days. Males accounted for less than 5 per cent of the
population, and lived 1.5 ± 0.1 days. Ghulam Abbas et al., (2010) showed, in laboratory conditions the average number of crawlers per batch averaged 20-53; a female of *P. solenopsis* could produce up to four batches of crawlers at 2-5 day intervals.

1.5 Research area IV:
Effect of recommended insecticides on the important predators and its prey *P. solenopsis*

1.5.1 Effect of insecticides on *Phenacoccus* spp.

Field trials conducted in Malawi to test the efficacy of fenitrothion, pirimiphos-methyl, dimethoate and fenvalerate for the control of *P. manihoti* showed that none of the treatments could increase yield (Anonymous 1988/89). Density of Japanese mealybug, *Planococcus kraunhiae* (Kuwana), on Japanese persimmon fruit was higher in plots frequently treated with cypermethrin than that in the untreated plot (Morishita, 2005). Populations of mealybug, *P. kraunhiae* were tested against insecticides by potter tower method in Japan, Neonicotinoids were the most toxic followed by organophosphates, while the synthetic pyrethroids were less effective (Morishita, 2006). Application of insecticides like dimethoate (0.05%) and monocrotophos (0.05%) failed to control the *F. virgata* on tuberose at the Indian Institute of Horticultural Research (IIHR), Bangalore (Mani and Krishnamoorthy 2007). Studies carried out in the laboratory of Pakistan, proved bifenthrin, profenofos and chlorpyrifos as best insecticides for *P. gossypiphilous* control based on its susceptibility with the leaf dip method (Shafqat et al., 2007). Suresh and Kavitha (2007) reported that imidacloprid, profenophos, acephate, chlorpyriphos and thiomethoxam were quite effective against *P. solenopsis* under the laboratory conditions but moderately effective under field conditions.

Dhawan et al., (2008) found endosulfan the least effective against *P. solenopsis*, collected from the cotton fields of Bathinda, Punjab. Ghulam, et al., (2009) proved that methidathion 40 EC, profenofos 50 EC, chlorpyrifos 40 EC and methomyl 40 SP were significantly effective against *P. solani* up to 7 days of treatment with mortality range of 85.74 to 95.69 per cent and 83.17 to 93.72 per cent during 2007 and 2006, respectively at Faisalabad, Pakistan. Out of 14 insecticides tested against *P. solenopsis*,
Kumar Rishi et al., (2009) found profenofos @ 1250 ml, monocrotophos @ 1250 ml, chlorpyriphos @ 3000 ml, quinalphos @ 2000 ml, acephate @ 2000 g, thiodicarb @ 625 g and carbaryl WP @ 2500 g/ha effective as spot sprays. Experiment, carried out on Bt cotton at Anand, Gujarat showed that carbaryl and chlorpyriphos rendered less than 70 per cent reduction of P. solenopsis population 3 days after spraying (Patel et al., 2010). Three days after spray carbaryl, profenophos, triazophos, methomyl and chlorpyriphos were all at par in reducing the population of P. solenopsis where per cent reduction varied between 67.93 to 75.36 (Jhala et al., 2010). Suresh et al., (2010) evaluated the insecticides by leaf dip method; chlorpyriphos recorded 100 per cent reduction of P. solenopsis followed by dichlorvos (90%) imdaclorprid (89.99%), thiamethoxam (86.66%) and profenophos (80%). However, when evaluated by insect dip method profenophos recorded the highest percentage of reduction (95.99) followed by endosulfan (94.96) and thiometoxam (93.41). Joshi et al., (2010) stated that, field sanitation, uprooting of infested plants and dusting of methyl parathion 2 per cent or spraying of profenophos 50 EC or chlorpyriphos 25 EC or quinalphos 25 EC help to reduce the population of P. solenopsis.

Bioefficacy of insecticides was evaluated against P. solenopsis at Anand (Gujarat), during year 2006-07. Out of nine different insecticides evaluated, profenophos (0.05 %), triazophos (0.04 %) and carbaryl (0.2 %) found to be the most effective for P. solenopsis in both laboratory and field conditions (Nikam et al., 2010b). Gulsar Banu et al., (2010) tested different treatments on P. solenopsis nymphs; acephate recorded the highest mortality of 53.3 per cent at 48h after treatment. Chlorpyriphos and Mealy Quit were equally toxic causing 48.9 per cent mortality at 48h after treatment. When tested against adults, the same trend of results was observed.

1.5.2 Effect of insecticides on Predators

Studies on the insecticides toxicity showed that dichlorvos 0.1% was safest insecticide whereas synthetic pyrethroids were highly toxic to adults of C. montrouzieri (Ramesh babu and Azam, 1987). Kalushkov (2000) studied the effect of permethrin, deltamethrin, fenpropathrin, esfenvalerate, alpha-cypermethrin and bioresemethrin on Coccinella septempunctata, Propylea quatuordecimpunctata, Adonia variegata
Hippodamia variegata] and Cycloneda limbifer. C. septempunctata had the highest survival rate. Thirumurugan and Gautam (2001) showed that the predators were more resistant to endosulfan than mealybugs. Rathore and Bapodra (2002) evaluated insecticides against the adult and grubs of C. septempunctata, Coccinella transversalis and Menochilus sexmaculatus in cotton at Junagadh, Gujarat. Endosulfan and dimethoate were found to be significantly safer followed by methyl-o-demeton, monocrotophos and phosalone. Cypermethrin and phosphamidon proved comparatively more toxic.

Experiment on okra in Orissa, revealed that predatory coccinellids population remained extremely low in malathion treated plot and were active in the biopesticide treated and untreated plots (Mishra and Mishra, 2002). Study at Andhra pradesh on okra showed that the neem formulations were relatively safe to M. sexmaculatus and Verania vineta as against monocrotophos (0.054%) and endosulfan (0.07%) (Gowri et al., 2002). Pasqualini and Civolani (2003) evaluated the effects of 6 insecticides on adults of the coccinellids Adalia bipunctata, C. septempunctata and Oenopia conglobata in apple, pear and peach orchards in Italy. Chlorpyrifos caused 40.2 and 63 per cent mortality in apples and peach respectively whereas malathion caused 43.5 per cent mortality. James (2003) studied the susceptibility of Stethorus punctum picipes and Harmonia axyridis grubs to insecticides. Chlorpyrifos and pirimicarb, produced 100 per cent mortality in S. p. picipes at concentrations equivalent to field rates. The insecticides, pirimicarb, endosulfan, and thiamethoxam were least toxic to H. axyridis. Bifenthrin, diazinon, dimethoate, methomyl, carbaryl, malathion, phosmet, imidacloprid, and chlorpyrifos were highly toxic. Sunitha et al., (2004) proved that dichlorvos and imidacloprid were toxic compared to their combination with eco-friendly chemicals against predatory coccinellid C. sexmaculata and Micraspis univittata on okra at Bapatla, Andhra Pradesh.

Al Doghairi, et al. (2004) studied the residual effects of chlorpyrifos (48 EC) and fenvalerate (20 EC) using panel exposure technique against A. variegata on Lucerne. chlorpyrifos was highly toxic to ladybird beetles at 1/2, 1, and 2 folds of the recommended dosage, where mortality percentages ranged from 46.7-50, 50, and 55-55.33 per cent, respectively. Fenvalerate produced moderate effect where mortality percentages were 10-47.7, 20-50, and 13.33-50 per cent, at 1/2, 1, and 2 folds. Field
investigations showed the insecticides alpha-cypermethrin and fipronil, caused significant decrease in the abundance of aphidophagous coccinellids *C. septempunctata*, *Coccinula quatuordecimpustulata*, *H. variegata* and *P. quatuordecimpunctata* (Kalushkov and Nedved 2005). Attia *et al.*, (2006) studied the effect of some alternative insecticides on coccinellid predators *Scymnus syriacus* and *Hyperaspis vinciguerrae* of *F. virgata*. Sumithion was harmful for adults of both the predators. Thornham *et al.*, (2007) studied the feeding responses of *C. septempunctata* to aphids contaminated with insecticides of class carbamate, organophosphate and pyrethroid. Consumption was reduced mostly in the pyrethroid treatment. Thornham *et al.*, (2008) studied the responses of insecticides belonging to different classes against Coccinellids. Pyrethroids significantly reduced locomotion. Organophosphates effected mixed locomotory responses and Carbamates effected very few changes in locomotory activity.

1.6 Research area V:
Effect of recommended insecticides on the solitary endo-parasitoids, of *P. solenopsis*.

1.6.1 Effect of insecticides on Parasitoids

Insecticides, such as endosulfan (0.07%), phosalone (0.07%) and dichlorvos (0.20%) showed less toxic residual activity while chlorpyrifos (0.05%), carbaryl (0.10%) and fenthion (0.10%) showed significantly more residual activity to both the encyrtid parasitoids, *Aenasius advena* and *Blepyrus insularis*, the key parasitoids of *F. virgata* (Mani, 1992). Field applications of chlorpyrifos, fenthion, methyl parathion [parathion-methyl] and quinalphos were toxic to pupae of parasitoids, *A. advena*, *Anagyrus qadrii* and *B. insularis*. Dimethoate and kerosene 2 per cent emulsion were less harmful (Balakrishnan *et al.*, 1991). The side effect of some pesticides was tested by Yigit *et al.*, (1992) on citrus mealybug parasitoid, *L. dactylopii*, in the laboratory which proved that, Chlorpyrifos-ethyl [chlorpyrifos], methidathion and fluvalinate to be harmful to adults of *L. dactylopii* and inhibited their emergence when applied to pupae.

Wakgari and Giliomee (2003) carried out laboratory bioassay of nine contact insecticides (methidathion, methomyl, methyl-parathion, parathion, profenofos and prothiofos) against mealybug parasitoid, *Coccidoxenoides peregrinus* (Hymenoptera:
Encyrtidae) which indicated that all were highly toxic, causing 98-100 per cent mortality in less than 6 hours of treatment. Attia et al., (2006) studied the effect of some alternative insecticides on the encyrtid parasitoid, *B. insularis* of *F. virgata*. Sumithion was found slightly harmful for the parasitoid.

### 1.7 Expected outcome

The present study shall generate the much needed information about biology, population dynamics, natural enemy complex and management tactics for solenopsis mealybug, pertaining to Indian conditions. Generated information in turn will help in devising ideal integrated pest management (IPM) strategy for this new menace threatening cultivation of field, fruit, vegetable, ornamental and medicinal crops.
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### 3.7 Predators of *P. solenopsis*

- a. *Hyperaspis maindroni*
- b. *Brumoides suturalis*
- c. *Scymnus coccivora*
- b. *Nephus regularis*

### 3.8 Predators of *P. solenopsis*

- a. *Coccinella septempunctata*
- b. *Cheilomenes sexmaculata*
- c. *Mallada* sp.
- b. *Chrysoperla carnea*

### 5.1 Rearing of *P. solenopsis*, on *Parthenium* stem cutting inside the homeopathic vial

### 5.2 Unmated female of *P. solenopsis*

- a) Produced the ovisac containing unfertilized eggs
- b) Ovisac containing the shells of unfertilized eggs, which did not yield nymphs

### 5.3 Life cycle of Solenopsis mealybug, *P. solenopsis*, on *Parthenium*

### 6.1 *Cryptolaemus* adult emerged from chlorpyriphos treated grubs (De-Melanization)

### 6.2 Phyto-toxicity of dichlorvos to China rose leaves
The present studies were carried out at 27±2°C temperature and 60±5% relative humidity (RH) in the Biological Control Laboratory (BCL), Division of Entomology, Indian Agricultural Research Institute, New Delhi, during the year 2007-09. The details of the rearing equipments, chemicals, the methodology adopted for carrying out various studies and statistical analysis used for interpretation of data are discussed briefly in this chapter.

2.1 Materials

2.1.1 Glass/Plasticwares

i. Plastic and glass jars (10 x 7 cm and 22.5 x 15 cm size) for rearing of *P. solenopsis* and its natural enemies (N.E’s).

ii. Glass cylinders (10 x 4 cm size) for rearing mummies of mealybug and bioassay studies of insecticides on mealybug and its N.E’s.

iii. Glass Homeopathic vials (7 x 1.5 cm size) for Biology studies on *P. solenopsis* and for rearing mummies of mealybug.

iv. Flask, measuring cylinders and micro pipettes of various sizes for preparation of the insecticidal solutions.

v. Reagent bottles (100 ml) for storing insecticide solutions.

vi. Glass Petri plates of 10 cm diameter for bioassay studies of insecticides on mealybug and its N.E’s.

vii. Plastic trays (30 x 10 cm and 15 x 7.5 cm size) for keeping the vials and the Petri plates, containing test materials.

2.1.2 Equipments

i. Binocular microscope with an incident light for observing different growth stages of the insects and N.E’.

ii. BOD incubator for maintaining the required temperature (27 ± 2°C) and humidity (60 ± 5%) conditions for optimum growth of the insects.
iii. Leica microscope (EZ-4) attached with the computer system having “Software” for photography of different stages of *P. solenopsis* and its predators/parasitoids.

iv. Glass atomizer for spraying the small quantity of insecticides for bioassay studies.

**2.1.3 Experimental material**

i. Sprouted potato tubers and tender parthenium stem cuttings as a substrate for rearing *P. solenopsis*.

ii. Nucleus culture of mealybug, *P. solenopsis* reared on sprouted potato tubers

iii. Nucleus cultures of coccinellid/chrysopid predators viz., *Hyperaspis maindroni*, *Cheilomenes sexmaculata*, *Coccinella septempunctata*, *Cryptolaemus montrouzieri*, *Nephus regularis*, *Scymnus coccivora* and *Mallada basalis*.

iv. Nucleus culture of parasitoid, *A. bambawalei* maintained on the *P. solenopsis*.

**2.1.4 Chemicals**

i. Formalin solution for disinfection of the potato tubers, rearing containers and glass wares in order to prevent any contamination due to fungal or bacterial infections.

ii. Ethyl alcohol for disinfection of hands and glassware’s for Bioassays.

iii. Ethyl alcohol for preservation of various stages of mealybug and its N.E’s.

iv. Ethyl alcohol, KOH, DPX mountant, Xylene, Glacial Acetic Acid, Carboxylol, Clove oil etc. for slide preparation of various stages of mealybugs and its N.E’s.

v. Insecticides viz.; chlorpyriphos (Dursban 20% EC), endosulfan (Endocel 35% EC), monocrotophos (Chetak 36% SL), malathion (Suryathon 50% EC), dichlorvos (Doom 76% EC) and alphamethrin (Alphagold 10% EC) for bioassay studies (Tab. 2.1).

vi. Acetone for preparing insecticide solutions.
2.1.5 Miscellaneous

i. Black muslin cloth and rubber bands to cover the rearing jars containing mealybug and its N.E’s.

ii. Cotton plugs, tissue papers, blotting papers etc.

iii. Needles, forceps, camel hair brush for handling stages of *P. solenopsis* and its N.E’s.

iv. Honey solution as a food for the adults of coccinellid predators and also for parasitoids of *P. solenopsis*.

v. Glass slides and cover slips for preparing slides of test insects and its N.E’s.

2.2 Methodology

2.2.1 Programme of research work

It consist of the following activities/milestones.

i. Survey of the N.E’s i.e. parasitoids and predators of *P. solenopsis* infesting different hosts.


iii. Rearing of *P. solenopsis* on a different collected host along with sprouted potatoes for emergence of N.E’s and maintaining the culture of different N.E’s.

iv. Examine the biology of *P. solenopsis* on host, *Parthenium hysterophorus*.

v. Elucidate the effect of insecticides against the *P. solenopsis* and its N.E’s (Predators and Parasitoids).

2.2.2 Survey of N.E’s of *P. solenopsis*.

Colonies of *P. solenopsis* infesting different hosts were collected and kept in plastic jars for rearing. The emerged N.E’s were then preserved in Ethyl alcohol (90%) for later taxonomic identification.

2.2.3 Population Dynamics of *P. Solenopsis* and its N.E’s.

Weekly observations of *P. Solenopsis* and its N.E’s were taken on its planted host such as brinjal, sorrel, okra and already planted host gurhal (Plate 2.1). The data
obtained was then subjected to the analysis of mean and its “Correlation Coefficients” with meteorological data were calculated.

2.2.4 Rearing of *P. solenopsis* on sprouted potato tubers

The mealybug culture was maintained in the laboratory by rearing on sprouted potato tubers (Plate 2.2) by following the method suggested by Gautam (1987, 2008b). Medium sized sprouted potato tubers (5-6) were kept in each plastic jar. Newly emerged crawlers from other maintained colonies or gravid females collected from field were released on the potato sprouts with the help of camel hair brush. The jars were covered with the double layer of clean black muslin cloth and tied with the rubber band firmly to prevent the escape of mealybugs. The jars were kept at 27 ± 2°C and 60 ± 5% RH. This mealybug culture was further used for future studies.

2.2.5 Maintaining coccinellids and chrysopids predator of *P. solenopsis*

The nucleus cultures of coccinellid predators were maintained on the mealybug in the laboratory by following the method suggested by Gautam (2008b). Five pairs of the predator were released in a jar containing sprouted potatoes infested with mealybug. The jars were covered with clean black muslin cloth and secured with the rubber band and then maintained at 27 ± 2°C and 60 ± 5% RH. Adults and grubs were used based on the availability for bioassay studies with the insecticides.

2.2.6 Maintaining Parasitoid of *P. solenopsis*

Encyrtid parasitoid, *A. bambawalei* was reared on *P. solenopsis* infesting sprouted potatoes in the laboratory at 27 ± 2°C and 60 ± 5% RH. Adults of the parasitoid were used for bioassay studies with the insecticides.

2.2.7 Preparation of Insecticide solution

Insecticidal solution of six insecticides viz; chlorpyriphos (Dursban 20% EC), endosulfan (Endocel 35% EC), monocrotophos (Chetak 36% SL), malathion (Suryathion 50% EC), dichlorvos (Doom 76% EC) and alphamethrin (Alphagold 10% EC) were made by dissolving required quantity of insecticides in solvent acetone for getting 0.05, 0.07, 0.04, 0.125, 0.15 and 0.01 % solution, respectively
(Tab. 2.1). Insecticidal solutions were used fresh and prepared as per the experimental requirement.

### 2.2.8 Preparation of stomach poison:

Honey (0.5 gm) was taken in six homeopathic glass vials and 1ml of each six insecticide solution prepared was added in respective vials. Then the vials were left overnight for evaporation of acetone, so that insecticide gets mixed with honey. Thus the concentrations of stomach poison prepared out of six insecticides were 0.10, 0.14, 0.08, 0.25, 0.30 and 0.02 %, respectively (Tab. 2.1). Stomach poisons were used fresh and prepared as per the experimental requirement.

### 2.2.9 Statistical analysis

Data related to the population dynamics of mealybug, *P. solenopsis* and its natural enemies were subjected to the correlation coefficient analysis by software “Microsoft office Excel-2007”. In order to know the interaction between treatments, data of insecticidal bioassays were subjected to two way factorial CRD analysis in SPSS statistical programme (Version 13.0) and the means were tabulated by Gomez and Gomez (1984). Before the statistical analysis per cent mortality data was transformed by arc-sine transformation. Critical difference (P) was worked out for testing the variance ratio is significant or not. The treatment effects were tested at 5 per cent probability level for significance.
Natural enemy complex of *Phenacoccus solenopsis* (Hemiptera: Sternorrhyncha: Coccoidea: Pseudococcidae), infesting different host’s plant.

### 3.1 Abstract and Keywords

Samples of an exotic mealybug, *P. solenopsis* collected from Delhi, Maharashtra and Andhra Pradesh during 2007 to 2010 were examined for the occurrence of its parasitoid and predators. A total of 17 parasitoids, belonging to seven families were found associated with various stages of *P. solenopsis*. The natural enemy complex comprising four primary parasitoids, eleven hyper-parasitoids as well as two unidentified chalcidoids were isolated from the samples. An encyrtid, *Aenasius bambawai*aei, was recorded as a main primary parasitoid of *P. solenopsis*, which served as primary host of all hyper-parasitoids. Among hyperparasitoids, *Promusciea unfasciativentris* was predominant. Interestingly, *Aphanogmus* sp. and *Anastatus* sp. are recorded for the first time on *P. solenopsis*. Besides, a total of ten predators *viz.*; eight coccinellids and two chrysopids were observed on *P. solenopsis*.

**Key words:** Host plants, hyper-parasitoid, *P. solenopsis*, *A. bambawai*aei, parasitoid, predator, survey

### 3.2 Introduction

The solenopsis mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Sternorrhyncha: Coccoidea: Pseudococcidae) has recently been reported from 35 global localities of various ecological zones so far attacking 183 plant species of 52 families (Ben-Dov *et al.*, 2009). The sap sucking mealybugs in general are very devastative in absence of their natural enemies besides difficult to control with the insecticidal applications while better amenable to biological control (Gautam, 2003). Many researchers in India and abroad time to time reported *P. solenopsis* as a new, exotic and invasive mealybug species (Gautam, 2007; Hodgson *et al.*, 2009; Ananthakrishnan and Jesudasan, 2009; Suresh *et al.*, 2010; Pala and Saini, 2010) with limited information on its natural enemy complex. Presuming that there could
be a number of local natural enemies having potential to develop appetite on this exotic mealybug for its suppression besides possibility of accidental entry of natural enemies along with the host, the present study was undertaken in Biological Control Laboratory, Division of Entomology, Indian Agricultural Research Institute, New Delhi, India. Keeping in view, the samples of the exotic mealybug, *P. solenopsis* collected from different ecosystems of Delhi, Maharashtra and Andhra Pradesh were examined for the occurrence of its parasitoid and predators during 2007 to 2010 besides in-depth experimentation selecting brinjal, china rose, sorrel and okra on experimental farm of the division.

3.3 Materials and Methods

3.3.1 Survey of *P. Solenopsis* natural enemies.

During the course of roving survey, different host plants heavily infested with *P. solenopsis*, were removed gently using secateur (4-5 branches) and brought to the laboratory and cultured at $27\pm2^\circ$C temperature and $60\pm5\%$ relative humidity on sprouted potatoes (Gautam, 2008b). Host plants collected were noted down and emerged natural enemies (parasitoids and predators) were preserved in 90 per cent ethyl alcohol. Predators associated with mealybug infested branches of respective host plants were also noted down in field itself, during roving survey. Samples of mealybugs, received from the state of Andhra Pradesh and Maharashtra, were also analysed same way for natural enemies.

In depth survey, selecting brinjal, china rose, sorrel and okra planted in experimental field of Division of Entomology, IARI, N. Delhi, was also done. Total of 5 plants of brinjal, 5 plants of sorrel, 20 plants of okra and 22 plants of China rose were surveyed weekly at random. Mummies, of *P. solenopsis*, were collected with the help of camel hairbrush and forceps; for brinjal from 3 branches (top, middle and lower), and sorrel from 5cm length of top of main stem, for okra 3 pods (top, middle and lower) for China rose from 10cm length of top of main stem per plant. Mummies were then brought to the laboratory, counted and kept individually in the numbered homeopathic vials for emergence of parasitoids. Observation on number of mummies collected, number kept for emergence and number of parasitoids emerged, per cent mortality and type of parasitoid emerged were noted down in detail.

Parasitoids were watched under Leica (EZ-4) microscope for taking photos and observing their body characteristics, in order to avoid mixing them with other
type of parasitoids and stored in 90 per cent ethyl alcohol. Parasitoids were identified and also preserved at Aligarh Muslim University (AMU), Aligarh, Uttar Pradesh; Prof. T. C. Narendran Trust for Animal Taxonomy, Calicut, Kerala and Insect Identification Service of Indian Agricultural Research Institute (IARI), Pusa, New Delhi. Status of the parasitoids was checked with the help of, Chalcidoidea (2011) and as per, Noyes and Hayat (1994). The predators collected from field as well as emerged from maintained culture of *P. solenopsis*, brought from different host plants, during roving survey were identified based on the literature and, Coccinellidae of the Indian subcontinent (2011).

3.4 Results

3.4.1 Survey of *P. Solenopsis* natural enemies.

Findings about the parasitoids complex of *P. solenopsis* are presented in the Table No. 3.1 and Plate No. 3.1, 3.2, 3.3, 3.4, 3.5, 3.6. Result indicate that in all, total of 17 parasitoids belonging to 7 families has been reported for *P. solenopsis*, which contains ‘4’ primary parasitoids (*Aenasius bambawalei* (Encyrtidae), *Leptomastix* sp. (Encyrtidae), *Paranathrix tachikawai* (Encyrtidae) and *Anagyrus* sp. (Encyrtidae)); ‘11’ hyper parasitoids (*Promusidea unfasciativentris* (Aphelinidae), *Marietta leopardina* (Aphelinidae), *Myiocnema comperei* (Aphelinidae), *Aprostocetus purpureus* (Eulophidae), *Prochiloneurus albifuniculus* (Encyrtidae), *Prochiloneurus testaceus* (Encyrtidae), *Prochiloneurus pulchellus* (Encyrtidae), *Cheiloneurus* sp., (Encyrtidae), *Chartocerus walkeri* (Signiphoridae), *Aphanogmus* sp. (Ceraphronidae) and *Anastatus sp.* (Eupelmidae)) and ‘2’ with unknown status (*Unknown encyrtid* (Encyrtidae) and *Unknown pteromalid* (Pteromalidae)) were isolated from samples drawn from Delhi, Mahaharashtra and Andhra Pradesh in India. Findings about the predator complex of the *P. solenopsis* are presented in the Table 3.6 and Plate 3.7, 3.8. Result indicate that in all, total of 10 predators belonging to 2 families has been reported for *P. solenopsis* which contains ‘8’ predators (*Hyperaspis maindroni*, *Brumoides suturalis*, *Scymnus coccivora*, *Nephus regularis*, *Coccinella septempunctata*, *Cheilomenes sexmaculata*, *Adonia variegata and Illeis* sp.) belonging to coccinellidae and ‘2’ predators (*Chrysoperla carnea* and *Mallada* sp.) belonging to chrysopidae.
Results, for in depth survey of four host of \textit{P. solenopsis} viz., brinjal, China rose, sorrel and okra are presented in Tab. 3.2, 3.3, 3.4, 3.5 and given as follows.

**Brinjal**

Total of 17 mummies, were kept for rearing and all emerged \{\textit{A. bambawalei} (3) and \textit{P. unfasciativentris} (14)}\}. Per cent mortality of mummies recorded was 52.78.

**China rose**

Total of 330 mummies were kept for rearing, 197 \{\textit{A. bambawalei} (16), \textit{P. unfasciativentris} (151), \textit{A. purpureus} (20), \textit{M. leopardina} (3), Unknown Pteromalid (1), \textit{Chartocerus walkeri} (3), \textit{Prochiloneurus albifuniculus} (3)}\} emerged. Per cent mortality of mummies recorded was 40.30.

**Sorrel**

Total of 144 mummies were kept for rearing, 129 \{\textit{A. bambawalei} (6), \textit{P. unfasciativentris} (122), \textit{Chieloneurus} (1)}\} emerged. Per cent mortality of mummies recorded was 40.30.

**Okra**

Total of 98 mummies were kept for rearing, 86 \{\textit{A. bambawalei} (47), \textit{P. unfasciativentris} (28), \textit{Chartocerus walkeri} (8), \textit{Prochiloneurus albifuniculus} (2), Unknown encyrtid (1)}\} emerged. Per cent mortality of mummies recorded was 12.15.

### 3.5 Discussion

#### 3.5.1 Survey of \textit{P. solenopsis} natural enemies.

All the four primary parasitoids of \textit{P. solenopsis}, belong to the family Encyrtidae only; the predominant species was \textit{A. bambawalei}. Reported 11 hyper-parasitoids belonged to the 6 families; the predominant species was \textit{P. unfasciativentris} followed by \textit{A. purpureus} and \textit{C. walkeri}. Among the 2 parasitoids whose status about parasitism could not be ascertained were a pteromalid and an encyrtid. Tanwar et al., (2008); Hayat, (2009); Gautam et al., (2009) and Vennila et al., (2010) already reported \textit{A. bambawalei}, as a key parasitoid of \textit{P. solenopsis}. \textit{P. tachikawai}, which was reared from the culture of \textit{P. solenopsis} collected from \textit{Parthenium hysterophorus}, is a gregarious parasitoid of very small size; it is in
conformity with Noyes and Hayat (1994). Species *Leptomastix* and *Anagyrus* are already reported as a primary parasitoid of many homopteran pests and mealybug as per Chalcidoidea, (2011) and Abd Rabou and Hendawy (2005). Among the hyper-parasitoids, *P. unfasciativentris*, *M. leopardina*, *M. comperei*, *A. purpureus*, *P. albifuniculus*, *P. testaceus*, *P. pulchellus*, *Cheiloneurus* sp. and *C. walker* are also reported as a hyper-parasitoids by many authors viz., Noyes and Hayat (1994); Abd Rabou and Hendawy (2005); Das and Sahoo (2005); Hayat et al., (2007); Vennila et al., (2010); Ram and Saini, 2010 and Chalcidoidea, (2011). *Aphanogmus* and *Anastatus* sp. are reported for the first time on mealybug, *P. solenopsis*. Both are presumed to be hyper-parasitoids. Gregory et al., (2005) already reported that, *Aphanogmus* species are usually parasitoids of cecidomyiid flies and are sometimes reared as hyper-parasitoids through hosts belonging to various insect orders. *Anastatus* sp. which looks like tiny ant is about 2mm long; they are all egg parasites of various insects including Lepidopterans, Orthopterans and Heteropterans, also known as secondary parasites; it is well supported by Eupelmid, (2011). Coccinellids were the predominant predators followed by the chrysopids. Unlike parasitoids, predators were reported from all the surveyed hosts of *P. solenopsis*. Many authors viz., Gautam et al., (2007); Ram and Saini (2010); Joshi et al., (2010); Fand et al., (2010) and Vennilla et al., (2010b) also reported important coccinellids and chrysopid predators of mealybug, *P. solenopsis*.

Discussion, about in depth survey of four hosts of *P. solenopsis* viz., brinjal, china rose, sorrel and okra are given as follows.

**Brinjal**

Brinjal recorded *A. bambawalei* and its parasitoid *P. unfasciativentris*, only. The number of mummies collected from brinjal were lowest among the four crops, may be due to brinjal being the less favoured host of *P. solenopsis*.

**China rose**

Among the four hosts under study, highest number of mummies of *P. solenopsis* were collected from china rose, due to it being the most favoured host; maximum diversity of parasitoids were also noticed in china rose. *M. leopardina* was recorded only from china rose, might be because of specific kairomone present in china rose. Abd Rabou and Hendawy (2005) also reported *M. leopardina* from pink hibiscus mealybug, *M. hirsutus*, a key pest of china rose. Per cent mortality of
mummies during August was more due to rainfall (299.1 mm), than during July (166.2 mm).

**Sorrel**

Number of mummies collected during October were highest than August and September because total rainfall received in October was nil, August had 299.1 mm, whereas September received 115.6 mm.

**Okra**

Third highest mummies were collected from okra and compared to other hosts recorded highest numbers of *A. bambawalei* than its hyper-parasitoid, *P. unfasciativentris*, an ideal condition for natural biological control.

### 3.5.2 Conclusion

Primary parasitoid, *A. bambawalei*, keeps population of *P. solenopsis* under check and 11 primary parasitoid of *A. bambawalei*, which are nothing but hyper-parasitoids of *P. solenopsis*, keeps population of *A. bambawalei* under check. The phenomenon of hyper-parasitism is not considered good in the biological control and 11 hyper-parasitoids are just too much for an efficient parasitoid, *A. bambawalei*. On a good part not a single hyper-parasitoid is gregarious but solitary; otherwise a successful fortuitous biological control of *P. solenopsis*, with *A. bambawalei*, would not be possible.
4. RESEARCH PAPER II

Population dynamics of exotic mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Sternorrhyncha: Coccoidea: Pseudococcidae) and its natural enemies associated with different plant hosts

4.1 Abstract and Keywords

Studies revealed that, Brinjal (var. shyamala) planted on 31\textsuperscript{st} July 2007 recorded highest (33.8 adults) *P. solenopsis*, in March 2008 and lowest (0.93 adults) in November 2007, mealybugs found positively correlated with temperature. However, when planted 14\textsuperscript{th} July 2008 remained free from the mealybugs except scanty population noticed during January 2009. The other variety of Brinjal (var. Vikram) planted on 14\textsuperscript{th} July 2008 was found negatively correlated with rainfall (-0.21). The parasitoid, *A. bambawalei*, was found negatively correlated with *P. solenopsis* (-0.21) and *P. unfasciativentris* (-0.12). Highest (53.73 adults) population of *P. solenopsis* was observed during November followed by October (21.13 adults) and lowest observed in December (1.3 adults) on sorrel planted in 2007. Highest (91.7 adults) population of *P. solenopsis* was observed during July followed by June (54.5 adults) and lowest was seen in December (1.3 adults) on sorrel planted in 2008.

Mealybug found positively correlated with temperature in sorrel planted 10.07.2007 and 15.04.2008 Highest number of *P. unfasciativentris* were recorded in October 2008 (116 adults) on sorrel and observed negatively correlated (-0.2) with mealybugs.

Okra planted 26.06.2008, recorded highest (11.08 adults) population of *P. solenopsis* during October as compared to lowest (3.4 adults) in November. It was positively correlated with temperature. Highest population of *A. bambawalei* was observed during October (23 adults), and found positively correlated with temperature (0.68) and *P. unfasciativentris* (0.11) which was highest in highest in November (26 adults).
China rose recorded highest (51.82 adults) population of *P. solenopsis* in July 2008, while lowest (0.06 adults) in October 2008. Mealybug was positively correlated with temperature (0.53), relative humidity (0.67) and rainfall (0.61). Mealybug, was again found positively correlated with parasitoid, *A. bambawalei* (0.70), hyper-parasitoid, *P. unfasciativentris* (0.97) and hyper-parasitoid, *A. purpureus* (0.73).

**Key Words:** Brinjal, China rose, natural enemies, Okra, *Phenacoccus solenopsis*, population dynamics, Sorrel

**4.2 Introduction**

Solenopsis mealybug, *Phenacoccus solenopsis* Tinsley, has been the current topic of research for insect taxonomists and applied entomologists in India due to its invasiveness, rapid spread, morphological and biological variations and the need for establishing an effective control strategy (Vennila *et al.*, 2010). *P. solenopsis* has been reported from 35 localities of various ecological zones across the globe (Ben Dov *et al.*, 2009). Survey across 47 locations of the country between months of late 2007 and early 2008 established the predominance of *P. solenopsis* in India (Nagrare *et al.*, 2009). By the end of the Kharif season (June–October), the total damage due to mealybug in 2007 was estimated to range from US $ 400,000 to 500,000 in north India alone. Apart from the yield losses, the pest infestation increased the cost of insecticide application by US $ 250–375 per acre in both India and Pakistan (Goswami, 2007). Of late, increased build up of various mealybug species such as *P. solenopsis* and *Paracoccus marginatus* in field and fruit crops is observed mainly due to certain abiotic changes in environment because of global warming. *P. solenopsis* has recently emerged as a serious pest in India, attacking important field crops such as cotton, okra, tomato, brinjal, chilli and ornamental plants such as China rose, particularly in nine cotton-growing states of India, Punjab, Haryana, Rajasthan, Gujarat, Madhya Pradesh, Maharashtra, Tamil Nadu, Andhra Pradesh and Karnataka (Dhawan *et al.*, 2007; Tanwar *et al.*, 2007; Jhala *et al.*, 2008; Dhara Jothi *et al.*, 2008 and Bhosle *et al.*, 2009).

Lot has been said about mealybug withstanding rising temperatures and getting washed away after rains (Valuli and Kosol, 1983; Dhawan *et al.* 2009), but still there is
scope for unraveling the role of weather factors on mealybug and its natural enemies, residing different crop ecosystems. So, the present studies were carried out to explore the reasons behind population fluctuation of *P. solenopsis* and its natural enemies in open field condition and to know round the year activity period of *P. solenopsis* and its natural enemies present in ecosystem of four crops viz., Brinjal (*Solanum melongena*), Sorrel (*Hibiscus sabdarifa*), Okra (*Abelmoschus esculentus*) and China rose (*Hibiscus rosa-sinensis*). This knowledge might help in devising the chemical control strategy containing the schedule of prophylactic as well as preventive measures for *P. solenopsis*. Brinjal, Jamaican sorrel and Okra were studied being the important vegetable crops grown in India throughout the calendar year and ornamental crop, China rose, was considered being very important host, acting as source of inoculum to crops of economic importance.

4.3 Materials and Methods

Population dynamics studies of Solenopsis mealybug, *P. solenopsis*, were carried out on four crops viz., Brinjal (*Solanum melongena*), Sorrel (*Hibiscus sabdarifa*), Okra (*Abelmoschus esculentus*) and China rose (*Hibiscus rosa-sinensis*) in Division of Entomology, IARI, New Delhi.

4.3.1 Brinjal

Two season data was recorded from brinjal planted in *Kharif* 2007 and 2008. Brinjal cultivars viz., Shyamala (long fruit) and Vikram (round fruit) were transplanted on 31.07.2007 and 14.07.2008, respectively. Var. Shyamala, did not attract *P. solenopsis* population considerably in 2007, hence was again transplanted in 2008 along with var. Vikram in anticipation of round fruit var. will attract more mealybugs. Three branches (top, middle and lower) per plant were observed for *P. solenopsis* and its natural enemies; total of five plants were selected weekly for observation. Observations were recorded from September 2007 to May 2008 for 2007 planted and September 2008 to May 2009 for 2008 planted brinjal.
4.3.2 Sorrel

Two season data was recorded from Sorrel planted in kharif 2007 and Summer 2008. The observations for the presence of mealybug and its natural enemies were recorded from top 5 cm of main stem per plant from randomly selected 5 plants of sorrel at weekly interval starting from September to December (2007) and June to December (2008) for Kharif and summer crops, respectively.

4.3.3 Okra

Observations were taken from September to December 2008 on okra crop (variety Rohini) planted on 26.06.2008 Twenty plants were selected weekly at random and 3 pods (top, middle and lower) per plant were observed for the presence of mealybug and its natural enemies.

4.3.4 China rose

China rose planted along the hedge rows around the Entomology Division of IARI were selected at random. At weekly intervals, top 10 cm of main stem per plant of twenty two plants was observed for the presence of mealybug and its natural enemies. Observations were noted from June 2008 to May 2009.

4.3.5 Observations and evaluation

Observations, for mealybug were taken as stated above. Predators per plant were observed and noted down in field itself. Regarding the parasitoids, mummies, of *P. solenopsis* per plant were collected with the help of camel hair brush and forceps, then brought to the laboratory, counted and kept individually in the numbered homeopathic vials for emergence of parasitoids. Observations on number of parasitoids and type of parasitoid emerged were noted down in detail. Means for population per plant were tabulated; correlation coefficients were calculated for mealybug, natural enemies and key meteorological parameters (average of minimum and maximum temperature, average of minimum and maximum relative humidity and rainfall per month) through ‘MS-Excel-2007’ for final interpretation of population dynamics data.
4.4 Results

4.4.1 Brinjal

Shyamala (2007): Results of var. shyamala for year 2007 are depicted in Fig. 4.1. Highest (33.8 adults) population of *P. solenopsis* was recorded during March 2008 and lowest (0.93 adults) in November. Month of December, January and February did not host mealybug, *P. solenopsis*. It was found positively correlated with temperature (0.20), negatively correlated with relative humidity (-0.41) and rainfall (-0.15). Natural enemies, *C. septempunctata* (0.47) and *C. sexmaculata* (0.17) were found positively correlated with mealybug. Other predators *B. suturalis* (-0.1) and *A. variegata* (-0.1) were found negatively correlated with mealybug.

Shyamala (2008): Results of var. Shyamala planted in 2008 are depicted in Fig. 4.1. *P. solenopsis* was not recorded during this period except in January 2009 (1 adult). Month of December, January and February did not host mealybug, *P. solenopsis*. Among natural enemies, only *C. septempunctata*, was noticed and found negatively correlated (-0.18) with mealybug.

Vikram (2008): Results of var. vikram planted in 2008 are depicted in Fig. 4.2, for which observations were recorded from September 2008 to May 2009. Maximum (3 adults) population of *P. solenopsis* was observed in November and rest of the months except February (0.5 adults) did not observed mealybug. Mealybug was found positively correlated with temperature (0.22), positively correlated with relative humidity (0.09) and negatively correlated with rainfall (-0.21). Parasitoid, *A. bambawalei*, was found negatively correlated with *P. solenopsis* (-0.21) and *P. unfasciativentris* (-0.12). However, *P. solenopsis* and *P. unfasciativentris* were positively correlated (0.97). Predators, *C. septempunctata* (-0.15) and *C. sexmaculata* (-0.03) were negatively correlated with mealybug.

4.4.2 Sorrel:

Results of sorrel planted in 2007 are depicted in Fig. 4.3 and 4.4. Highest (53.73 adults) population of *P. solenopsis* was observed during November followed by October (21.13 adults) and lowest in December (1.3 adults). Mealybug was positively correlated
with temperature (0.08) and negatively correlated with both relative humidity (-0.04) and rainfall (-0.26). Natural enemies were not observed on sorrel planted in 2007.

Results of sorrel planted in 2008 are depicted in Fig. 4.3 and 4.4. Highest (91.7 adults) population of *P. solenopsis* was observed during July followed by June (54.5) and lowest (1.3 adults) in December. Mealybug was found positively correlated with all three weather parameters viz., temperature (0.61), relative humidity (0.52) and rainfall (0.38). With respect to parasitoid, *A. bambawalei*, was highest (3 adults) in August, followed by September (2 adults) and lowest in October (1 adults), while other months did not record it. It was found negatively correlated (-0.34) with mealybug, whereas positively correlated with temperature (0.39), relative humidity (0.33) and rainfall (0.67). It was partially non-correlated with hyperparasitoid, *P. unfasciativentris* (0.08).

Highest (116 adults) number of *P. unfasciativentris* were recorded in October, followed by November (5 adults), lowest in September (1 adults) and other months did not record *P. unfasciativentris*. It was observed negatively correlated (-0.2) with mealybug, *P. solenopsis* and partially non-correlated with temperature (0.02). The other hyperparasitoid observed was *cheiloneurus* sp. (1) in October.

**4.4.3 Okra**

Results of Okra planted in 2008 are depicted in Fig. 4.5. Highest (11.08 adults) population of *P. solenopsis* was recorded during the October followed by September (5.05 adults), lowest during November (3.4 adults) and did not record any in December. It was found positively correlated with temperature (0.75), negatively correlated with relative humidity (-0.72), not correlated with rainfall (0.02) and positively correlated with *A. bambawalei*. Highest (23 adults) population of *A. bambawalei* was observed during October followed by November (13 adults) lowest in September (11 adults) and no record in December. It was found positively correlated with temperature (0.68), negatively correlated with relative humidity (-0.82), not correlated with rainfall (-0.05) and positively correlated with *P. unfasciativentris* (0.11). Hyper-parasitoid, *P. unfasciativentris*, was highest (26 adults) in November, lowest (1 adults) during September and October (1 adults) while December did not report it. Hyper-parasitoid, *Chartocerus walkeri*, was found non-correlated (-0.07) with mealybug and positively
correlated with its primary host *A. bambawalei*. Other parasitoids observed in less numbers were *Prochiloneurus albifuniculus*, and an unknown encyrtid.

### 4.4.4 China rose

Results of China rose for year 2008-09 are depicted in Fig. 4.6. Highest (51.82 adults) population of *P. solenopsis* was observed during July followed by August (22.01 adults) and June (6.29 adults), while lowest (0.06 adults) in October. From November to May, mealybugs were not observed. Mealybug was positively correlated with temperature (0.53), relative humidity (0.67) and rainfall (0.61). Mealybug, was again found positively correlated with parasitoid, *A. bambawalei* (0.70), hyper-parasitoid, *P. unfasciiventris* (0.97) and hyper-parasitoid, *A. purpureus* (0.73). Highest (6 adults) numbers of *A. bambawalei* were observed in August followed by June (4 adults), July (4 adults) and lowest in October (2 adults). January to May 2009 did not see any *A. bambawalei*. It was found positively correlated with temperature (0.72), relative humidity (0.71), rainfall (0.80), *P. unfasciiventris* (0.46) and *A. purpureus* (0.82). Highest (124 adults) *P. unfasciiventris*, were seen in July followed by August (27 adults) and other months did not record it. It was seen positively correlated with temperature (0.43), relative humidity (0.53) and rainfall (0.45). Other hyper-parasitoids, recorded from July to October were *Chartocerus walkeri* (3 adults), *P. albifuniculus* (3 adults), and *Marietta leopardina* (3 adults).

### 4.5 Discussion

#### 4.5.1 Brinjal

Mealybugs are known to proliferate if subject to increasing temperature; present findings showed positive correlation of mealybug population on var. shyamala (2007) with temperature and results are in conformity with Dhawan *et al.*, (2009). However, mealybug was found negatively correlated with both relative humidity (RH) and rainfall. It supported mealybugs getting washed off due to rains and it is well supported by Valuli and Kosol, (1983) who reported reduction in mealybug population in rainy season and by. Jayakumar *et al.*, (2009) also reported positive correlation of mealybug and RH, which is not proved in present findings. Besides, results indicate that mealybugs could
Mealybug population was at minimum during 2008 on var. shyamala, might be due to high rainfall in August 2008 (299.1 mm). Predator, *C. septempunctata* was negatively correlated with mealybug, which can not be explained as population of mealybug was at very minimum and found feeding on aphids in absence of mealybugs.

Mealybugs on var. vikram (2008) were again found positive and rainfall negatively correlated. It inferred that heavy rainfall of 2008 and probably brinjal not a favored host kept population of mealybugs at minimum. Mealybug, were found negatively correlated with parasitoid, *A. bambawalei*, hyper-parasitoid, *P. unfasciiventris* and two predators which might be due to minimum population of *P. solenopsis* and in its absence predators were feeding on aphids.

### 4.5.2 Sorrel

Again mealybugs on Sorrel (2007) were positively correlated with temperature and negative with RH and rainfall, it is in agreement with Valuli and Kosol, (1983) and Dhawan *et al.*, (2009) which is already discussed as above.

All three parameters (Temp., RH and Rainfall) were found positively correlated with mealybug. Findings related to the RH and mealybugs on sorrel (2008) are in agreement with Jeyakumar *et al.*, (2009) who also reported negative correlation of RH with mealybug intensity on cotton. Sorrel, which is also a malvaceae crop like cotton might be supporting mealybug buildup because of nutritional changes in crop due to low relative humidity. *A. bambawalei* and *P. unfasciiventris* was negatively correlated with mealybug which is understandable for Host: Pest association and positively with temperature which is well supported by Hanchinal *et al.*, (2010).

### 4.5.3 Okra

Mealybug population remained at peak during October and started declining from the onset of winter. As reported earlier with other crops, mealybugs were positively correlated with temperature which is in agreement with Hanchinal *et al.*, (2010), who reported on cotton, a malvaceae crop like okra. Mealybugs were positively...
correlated with its parasitoid, *A. bambawalei* and *A. bambawalei* was positively correlated with its parasitoid, *P. unfasciativentris*, an ideal situation in biological pest suppression; a phenomenon where as host population increases its natural enemy also increases and brings its host pest down to general equilibrium position (GEP).

### 4.5.4 China rose

Highest (51.81 adults) mealybugs of July were brought down in August (22.01 adults) because of heavy rains of August (299.1 mm). Mealybugs were positively correlated with temperature; it is well supported by Hanchinal *et al.*, (2010) and Dhawan *et al.*, (2009), but also reported negative correlation with RH and rainfall which is not reported in present findings. As in case of okra, mealybugs were positively correlated with *A. bambawalei* and *P. unfasciativentris*, an ideal situation for restoring general equilibrium position of mealybugs and its natural enemies. *A. bambawalei* was positively correlated with temperature also reported by Hanchinal *et al.*, (2010). *P. unfasciativentris* seen most active in July and August and not in other months, it is in agreement with Ram Pala and Saini (2010), who reported that during July-August *A. bambawalei* was attacked by hyper-parasitoids.

### 4.6 Conclusion

Brinjal a favored host of Solanum mealybug, *P. solani*, is not found a favored host of *P. solenopsis*. Crops belonging to Malvaceae family attracted considerable number of *P. solenopsis* and its parasitoids. Brinjal, Sorrel and China rose registered highest number of *P. unfasciativentris* than *A. bambawalei*, but in Okra, *A. bambawalei* over-numbered hyper-parasitoid, *P. unfasciativentris*. The situation in Okra is found to be most desirable for successful biological pest suppression. Unlike, in Brinjal and Sorrel, *A. bambawalei* and *P. unfasciativentris* were found restoring population of *P. solenops* at ‘General Equilibrium Position’ (GEP) in different crop ecosystems. The studies are in conformity with the findings of Gautam *et al.*, (2009) who reported the occurrence of the phenomenon of Fortuitous Biological Control and also supported by Ram Pala and Saini, (2010).
5. RESEARCH PAPER III

Bio-ethological studies on exotic mealybug, *Phenacoccus solenopsis* Tinsley in laboratory

3.1 Abstract and Keywords

Bio-ethology studies were carried out on *Phenacoccus solenopsis* in Biological Control Laboratory, IARI, New Delhi at 27 ± 2°C temperature and 60 ± 5% RH. Result showed that males went through 4 nymphal instars viz., I<sup>st</sup> (5.86 ± 0.82 days), II<sup>nd</sup> (5.5 ± 0.56 days), III<sup>rd</sup> (2.2 ± 0.41 days) and IV<sup>th</sup> (2.26 ± 0.45 days) as against the 3 instars in females I<sup>st</sup> (5.94 ± 0.82 days), II<sup>nd</sup> (3.70 ± 1.21 days) and III<sup>rd</sup> (4.41 ± 0.50 days). The total life cycle for female and male was noticed as 14.06 ± 1.09 & 15.53 ± 0.83 days, respectively. Pre-mating, pre-ovipositional, ovipositional and post ovipositional periods were recorded as 2.11 ± 1.26, 7.66 ± 0.70, 10.22 ± 2.68 and 3.77 ± 1.71 days, respectively. The adult longevity was reported as 47.125 ± 7.01 for unmated females, 3.66 ± 0.81 for unmated males, 24.66 ± 4.5 for mated females and 1.33 ± 0.50 days for mated males. Females once mate did not mate again. Reproduction was sexual and ovoviviparous, wherein only mated females laid eggs (167.77 ± 25.57/female); unmated females did not lay eggs. Life span (Life cycle duration and Adult longevity) was noticed as 61.5 ± 6.95, 18.66 ± 0.82, 38.44 ± 4.95 and 17.13 ± 0.97 days for unmated females, unmated males, mated females and mated males, respectively.

Key Words: Bio-ethology, life cycle, *Parthenium, Phenacoccus solenopsis*

3.2 Introduction

*Solenopsis* mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae) is a pest of ornaments and fruits trees worldwide and known to be cryptic in nature. It was described originally from the U.S. in 1898 and it remained known only in the U.S., where it is widespread, until 1992 (Ben-Dov 2004). Recently, in India *P. solenopsis* and *Phenacoccus solani* Ferris (Hemiptera: Pseudococcidae) were observed on several plant hosts including cotton (Jhala, *et al.* 2008). *Solanum* mealybug,
*P. solani* was reported attacking 64 plant species belonging to 27 families, including weeds in and around the Delhi (Gautam, 2007). To combat the pest menace pesticides of worth over Rs. 500 crores were sold in Punjab (India) in just three months from June to August 2007 (Dutt, 2007).

The mealybug species (*P. solenopsis*) is reported from variety of crops i.e. agricultural, horticultural, medicinal and ornamentals in different parts of country; present studies were carried out to have knowledge about its bio-ecology and weak links in life cycle of mealybug. *Parthenium* stem cuttings were used for study because; it is one of the very important hosts of *P. solenopsis* (Gautam, 2007), is available in plenty through out the year, easy to handle and it has a number of furrows on it wherein nymph settles firmly and does not fall or get disturbed while handling.

### 3.3 Materials and Methods

#### 3.3.1 Rearing of *P. solenopsis* on *Parthenium* Stem cuttings.

The studies on biology of *P. solenopsis* were carried out at 27 ± 2°C temperature and 60 ± 5% relative humidity (R.H) in the Biological Control Laboratory, Division of Entomology, Indian Agricultural Research Institute (IARI), New Delhi during the year 2007-09. Adult female was collected from the *Parthenium* plant from the field and the forty freshly hatched nymphs (< than 24 hrs.) were kept for rearing on *Parthenium* tender stem cuttings (4 cm long) of pencil size thickness in the individual Homeopathic vials (7 X 1.5 cm size) (Plate 5.1). First crawler was transferred by hair brush on to the stem cuttings and then stem cuttings were kept individually in the vials. The mouth of the homeopathic vials were secured with the paraffin wax film to prevent crawlers from coming out then paraffin wax film was punctured with entomological pins for making minute holes for crawlers breathing. Once the crawlers got settled next day paraffin wax film was replaced with cotton swab. The stem cuttings were changed daily by keeping it in the vials using forceps. As and when the mealybugs crawled on to the new stem cutting the older cutting was removed. For mating newly emerged male was released into the vial having grown up female. After mating the female was removed from the individual vial and kept in the Petri dish having circular shape black blotting paper for
anchorage; it also increased the visibility of minute eggs and just hatched crawlers while counting. The experiment had 40 replicates. The stages of the mealybug were cleared with the clove oil, before taking the photos with “Leica EZ-4” microscope attached to the computer.

3.3.2 Experimental design and evaluation

A homeopathic vial each containing *P. hysterophorus* stem cutting and a nymph was kept in 40 replications where each nymph was considered as one replicates. This experiment was again repeated with a set of forty (40) nymphs. Only those nymphs, which could complete full development, were assessed for the results. The biological parameters i.e. date of molting, nymphal duration (days), pupal duration (days), fecundity (number of eggs or crawlers/female), adult longevity (days), sex ratio (male: female) and life span (days) were noted during the course of study. Besides observations on behavior of mated and unmated male/female was also recorded. Generated data was subjected to simple mean and standard deviation in “Microsoft office 2007” for interpretation. Comparison of biological attributes of unmated/mated male and female was done by subjecting the data to paired t-test in “SAS Enterprise guide-4.2”, for comparing the differences in means. Sex ratio (Male: Female) was calculated based on the number of male and females that could complete development.

3.4 Results

Result are given as follows, except the result of paired t-test performed for comparing the biological attributes of unmated/mated males and females, which found non significant (Table 5.2).

3.4.1 Life cycle (Nymphal & Pupal development)

The results about the biology of *P. solenopsis* are depicted in the Table 5.1 and Plate 5.3. Male nymphal duration took 5.86 ± 0.35, 5.2 ± 0.56, 2.2 ± 0.41 and 2.26 ± 0.45 days for I, II, III (Pre-pupa) & IV (Pupa) instars, respectively; while female took 5.94 ± 0.82, 3.70 ± 1.21 & 4.41 ± 0.50 days for I, II & III instars, respectively. Male nymphs took 4.46 ± 0.63 days for total pupal development. The total developmental
period from egg to adult for female and male was noticed to be 14.06 ± 1.09 and 15.53 ± 0.83 days, respectively.

3.4.2 Adult longevity

It is the period from adult emergence up to its death. Unmated females took 47.125 ± 7.01 & unmated males 3.66 ± 0.81 days for adult longevity. However, the mated females took 24.66 ± 4.5 & mated males 1.33 ± 0.50 days for adult longevity. The adult longevity of mated female was again sub-divided into pre-mating, pre-ovipositional, ovipositional and post ovipositional period which were 2.11 ± 1.26, 7.66 ± 0.70, 10.22 ± 2.68 and 3.77 ± 1.71 days, respectively.

3.4.3 Fecundity

Female once mated did not mate again with the same and with other males. Only mated females could lay eggs at the rate of 167.77 ± 25.57 in 10.22 ± 2.68 days of ovipositional period. Unmated females could only construct ovisac, but did not lay eggs. Some females constructed ovisac and laid unfertile eggs, which hatched but did not yield nymphs. (Plate 5.2)

3.4.4 Life span

It is counted as a period of life cycle and adult longevity (period of hatching from egg to the death of adult). Unmated and mated females took 61.5 ± 6.95 and 38.44 ± 4.95 days, respectively. However, unmated males took 18.66 ± 0.82 and mated males took 17.13 ± 0.97 days towards the life span. Out of an eighty crawlers taken for study only 32 (40 %) could grow up to the adulthood and rest forty eight (60 %) succumb to death.

3.4.5 Sex Ratio

In total, seventeen (17) nymphs completed development into female and fifteen (15) nymphs did become males. So, the sex ratio for male: female, was identified as 1: 1.13, which means for every 100 males there were 113 females.
3.5 Discussion

Comparison between the biological attributes of pair of unmated/mated males and females, which found to be non significant, might be because most of the biological parameters pair of unmated/mated males and females share are common. Except adult longevity and life span other parameters remained same for unmated and mated males, while except adult longevity, fecundity and life span other attributes remained same for unmated and mated females. Discussion based on the mean and standard deviation is given as under.

3.5.1 Life cycle (Nymphal & Pupal development)

Experimental findings showed that females took 3 instars to become adult, whereas males took 4 instars. The total nymphaal duration for female and male was 14.06 ± 1.09 days and 15.53 ± 0.83 days, respectively. Sinacori, (1995) also reported female nymphaal instar (I, II & III) duration of 16.8 days for mealybug, *P. madeirensis*. It indicates that females become an adult 1.5 days prior to males. It is justifiable, since male had to pass through an extra moulting and adult females did mate 2.11 ± 1.26 days (Pre-mating period) after becoming adult. Correa *et al.*, (2005) for *Planococcus citri*, Nikam *et al.*, (2010) and Vennila *et al.*, (2010a) for *P. solenopsis*, reported longer male nymphaal period than females as the male has to pass through one extra pupal instar.

3.5.2 Adult longevity

Condition of mating and unmating for males and females revealed that, females lived longer than males for both. It is in agreement with Juang Horng Chong *et al.*, (2003), who reported adult longevity for *Phenacoccus madeirensis*, as 3 and 20 days for males and ovipositing females, respectively. The current, findings about the pre-ovipositional (7.66 ± 0.70) and ovipositional period (10.22 ± 2.68 days) nearly support the findings of Nikam *et al.*, (2010), who reported it as 8.56 ± 0.61 and 16.73 ± 0.57 days, respectively on *P. solenopsis*. Fand *et al.*, (2010a) also reported ovipositional period of 11.75 ± 0.96 days for *P. solenopsis*. Unmated males lived longer because of being less active and unmated females lived longer might be due to not laying eggs; thus utilizing stored food meant for egg development for her nutrition.
3.5.3 Fecundity

Mode of reproduction was sexual and ovoviviparous. Lloyd (1952) also reported reproduction in pseudococcidae being predominantly, bisexual. Since the female laid eggs after mating only it’s a sexual reproduction and ovoviviparous because the eggs hatched within few hours after laying, it means nymphs had already completed development inside the eggs retained by female. Vennila et al., (2010) also reported the ovoviviparous mode of reproduction in P. solenopsis. However evidence of their reporting with regards to progeny produced by unmated female (parthenogenesis) was not proved in the present investigation. Present findings of fecundity (167.77 ± 25.57 eggs/nymphs) are nearly in line with the findings of Gautam et al., (2007) who reported fecundity of P. solani to be 191 eggs on Parthenium weed.

3.5.4 Life span

Result about the life span indicates that, unmated individuals lived longer than that of mated and females lived longer than males. In case of unmated females, food stocks which they should reserved for the egg laying might be helping them live longer than mated females. Male life span was shorter than that of the female as it had extra pre-ovipositional, ovipositional and post ovipositional duration. Strickland (1951) observed that one virgin female of cacao mealybug, Planococcoides njalensis, lived 65 days after maturity without producing eggs. Present studies also reported a life span of 76 days for one unmated female without producing fertilized eggs (Plate 5.2).

3.5.5 Sex Ratio

Sex ratio for male: female, was identified as 1: 1.13, which means for every 100 males there were 113 females. This sex ratio indicates that, there is a male for every female, which is ideal for the insect sexual reproduction. Sex ratio identified for P. solenopsis is nearly in conformity with the Mukhopadhyay and Mukherjee (2005) who found male: female ratio to be 1: 1.30 for mealybug, Planococcus cajani, reproducing sexually and oviparously.
3.6 Conclusion

Studies revealed that, parthenium stem cuttings are ideal for studying the biology; as it is available throughout the year and easy to handle in the laboratory. Studies also showed that *P. solenopsis* females reproduced sexually and not parthenogenetically, so can easily manage through biological means. Past experience also tells about mealybugs reproducing parthenogenetically create havoc because it can multiply exponentially during unfavorable conditions and surpass the population of its key mortality factors (Parasitoids and Predators).
6. RESEARCH PAPER IV

Effect of recommended insecticides on *Phenacoccus solenopsis* Tinsley (Hemiptera: Sternorrhyncha: Coccoidea: Pseudococcidae), and its important coccinellid predators

6.1 Abstract and Keywords

Six insecticides *viz;* chlorpyriphos 20 EC (0.05 %), endosulfan 35 EC (0.07 %), monocrotophos 36 SL (0.04 %), malathion 50 EC (0.12 %), dichlorvos 76 EC (0.15 %), and alphamethrin 10 EC (0.01 %) were tested for their residual toxicity against the mealybug, *Phenacoccus solenopsis* and its coccinellid predators. Studies revealed that, both chlorpyriphos and malathion showed highest toxicity in terms of mean mortality (100 %) to female mealybugs at 24 hrs of exposure as against lowest in endosulfan (35 %). Interestingly, chlorpyriphos and dichlorvos, which proved toxic to mealy bug were less toxic to the grubs of *C. montrouzieri*, registering only 21.66 per cent and 34.16 per cent mortality, respectively. These insecticides when offered to the adults along with honey differed in toxicity as stomach poison at 24 hrs of exposure. Endosulfan registered mortality to the tune of 60 per cent and 70 per cent against *C. septempunctata* and *C. sexmaculata*, respectively. However, stomach toxicity pertaining to the insecticide-sprayed mealybugs as prey offered to all the three species of predators indicated that the insecticides, chlorpyriphos and endosulfan were lesser toxic to these wherein mean mortality ranged from 38.09 to 56.66 and 50 to 65.71 per cent, respectively. Dichlorvos was the most toxic registering 100 per cent mortality for all 3 predators *viz;* *N. regularis*, *S. coccivora* and *H. maindroni*.

Chlorpyriphos being also a fumigant in action probably was found selective and safer to *Cryptolaemus* and warrants further investigations. Besides, reasons, behind the de-melanization of adult *Cryptolaemus* emerged from chlorpyriphos treated grubs also need to ponder upon. It may be noted that release of *Cryptolaemus* coupled with chlorpyriphos is likely to be a better option for the management of mealybugs infesting several horticultural and other crop plants.
**Key Words:** Efficacy, insecticides, *Phenacoccus solenopsis*, safety evaluation, predators

### 6.2 Introduction

Mealybugs (Hemiptera: Pseudococcidae) are cottony in appearance, small oval, soft-bodied sucking insects and are sometimes mistaken for soft bodied scale insects (Gautam, 2010). Adults are found on leaves, stems and roots and are covered with white mealy wax, which makes it difficult to eradicate them (Osborne, 2011). More than 100 species of mealybugs are reported to attack a variety of plant species in India (Varshney, 1985). The exotic Solenopsis mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Sternorrhyncha: Coccioidea: Pseudococcidae), earlier reported as Solanum mealybug, *Phenacoccus solani* Ferris (Gautam, 2007; Gautam *et al*., 2007; Suresh and Kavitha, 2007; Gautam, 2008) was later reported as *P. solenopsis* (Thomas and Ramamurthy, 2008). It has caused havoc among the crops of economic importance in some parts of the country and warrants search for efficient bio-intensive management options including judicious use of chemical insecticides. The ineffectiveness of chemical insecticides against mealybug due to the presence of a waxy coating and non-availability of suitable alternative control options limit their effective management (Mohan *et al*., 2004). It could be achieved by the judicious use of pesticides, which could kill the pest effectively and remain less harmful to the natural enemies.

Good number of natural enemies comprising coccinellids *viz.*, *Hyperaspis maindroni* (*Bromoides lineatus*), *Bromoides suturalis*, *Cheilomenes sexmaculata*, *Cryptolaemus montrouzieri*, *Nephus regularis* and *Scymnus coccivora*; chrysopids, *Chrysoperla carnea* and *Mallada* sp. are known to feed on *Phenacoccus* spp. (Gautam *et al*., 2007; Sudhida *et al*., 2008, 2009; Vennilla *et al*., 2010). Before the inclusion of insecticide in integrated pest management modules or advocating it for chemical control independently, insecticide has to be tested for its bio-efficacy and safety against the mealybug and its predators, respectively. The present studies were carried out to elucidate the bio-efficacy of recommended insecticides against *P. solenopsis* and susceptibility of coccinellid predators of solenopsis mealybug, *P. solenopsis* to recommended insecticides.
6.3 Materials and Methods

Details of the insecticides used for bio-efficacy studies against *P. solenopsis* and for safety evaluation on predators are given in Tab. 2.1 in chapter 2.

6.3.1. Residual toxicity of insecticides against the females and crawlers of *P. solenopsis*

Six insecticides, *viz*; chlorpyriphos (0.05 %), endosulfan (0.07 %), monocrotophos (0.04 %), malathion (0.125 %), dichlorvos (0.15 %), and alphamethrin (0.01 %) were tested using field recommended doses against the female and crawler of *P. solenopsis*. Glass Petri plates (10 cm dia) for females and glass cylinders (10 x 4 cm size) for nymphs were used in experimentation. The inner sides of the plate and cylinders were smeared with 1 ml solution of respective insecticide in acetone (see section 2.2.7, in chapter 2). A batch of Petri plates/glass cylinders coated with acetone only was kept as control and the experiment was replicated thrice. Then, the Petri plates/glass cylinders were shade dried for half hour. After shade drying, a batch of fifteen females and twenty crawlers was released in to the Petri plates and glass cylinders, respectively. Females were provided with China rose leaves for feeding. The observations on mortality were recorded at 24 hr, 48 hr, 72 hr and 96 hr of exposure to the insecticides for the females. Since, the life of crawlers is not more than a day mortality data for it was recorded at 1 hr, 2 hr, 3 hr, 4 hr, 5 hr, 6 hr and 24 hrs of exposure to the insecticides.

In case of nymphs instead of Petri plates, glass cylinders were used because the crawlers being small in size were prone to escape from the Petri plates. The mouth of the glass cylinders was secured with paraffin wax film and punctured with entomological pins to facilitate crawler’s respiration and also to prevent their outward movement.

6.3.2. Residual toxicity of insecticides against the predators of *P. solenopsis*

Six insecticides *viz*; chlorpyriphos (0.05 %), endosulfan (0.07 %), monocrotophos (0.04 %), malathion (0.125 %), dichlorvos (0.15 %), and alphamethrin (0.01 %) at field dose were tested against the adults and grubs of predators namely *C. setempunctata*, *C. sexmaculata*, *N. regularis*, *S. coccivora* and *H. maindroni* and only grubs of *C. montrouzieri* and *Mallada* sp. Grubs used for studies were of last instar except for 2nd instar of *Mallada* sp. Petri plates (10×4 cm
size) were used for adult and grubs of all predators except *N. regularis* and *Scymnus coccivora* for which glass cylinders were used. Procedure of treating the Petri plates and glass cylinders remains same as discussed above. Experiment was replicated thrice. After shade drying the Petri plates and glass cylinders, a batch of (10 each adults/grub (24 hrs old) of *C. septempunctata* and *C. sexmaculata*; 15 each adults/grubs of *N. regularis* and *S. coccivora*; 10 each adults/grubs of *H. maindroni* and 10 grubs each of *C. montrouzieri* and *Mallada* sp. was released. The mouth of the glass cylinders was secured with black muslin cloth and tied with the rubber band. Live adults/grubs of predators after 24 hrs were transferred to new fresh Petri plates/glass cylinders and fed with mealybugs. The observations about the mortality were taken at 24 hrs, 48 hrs, 72 hrs and 96 hrs of exposure to insecticides.

6.3.3. Toxicity of insecticides incorporated honey (stomach poison) against the predators

Six insecticides viz; chlorpyriphos (0.1 %), endosulfan (0.14 %), monocrotophos (0.08 %), malathion (0.25 %), dichlorvos (0.3 %), and alphamethrin (0.02 %) were tested for stomach toxicity against the adults of two coccinellid predators viz; *C. septempunctata* and *C. sexmaculata*. For stomach toxicity studies, stomach poisons were made out of respective insecticide and honey (see section 2.2.8, in chapter 2). Petri plates (10 cm Ø) were taken and batches of 10 (24 hrs old) well-fed adult predators were released in to the Petri plates. The inner walls of Petri plates were smeared with respective stomach poisons. Batch of glass cylinders smeared with acetone treated honey were kept as control and the experiment was replicated thrice. After completion of 24 hrs, live predators were transferred to the fresh untreated Petri plates and fed with honey. The observations about the mortality were taken at 24 hrs, 48 hrs, 72 hrs and 96 hrs of exposure to insecticides.

6.3.4. Stomach toxicity of insecticides treated *P. solenopsis* against the predators

Six insecticides viz; chlorpyriphos (0.05 %), endosulfan (0.07 %), monocrotophos (0.04 %), malathion (0.125 %), dichlorvos (0.15 %), and alphamethrin (0.01 %) at field dose were tested against the adults of predators viz; *N. regularis*, *S. coccivora* and *H. maindroni*. A total 30 mealybugs consisting of different instars were kept in 7 glass cylinders (10 cm × 4 cm) and sprayed with 1ml of each 6 insecticide solution made in acetone 1ml acetone, along with control (only
acetone). All the treatments were sprayed with the help of hand glass atomizer. Treated glass cylinders were shade dried for half an hour. Then, treated mealybugs were removed and offered for consumption to the starving coccinellids kept in separate individual glass cylinders. Experiment was replicated thrice. After completion of 6 hrs, live predators were transferred to the fresh untreated glass cylinders and fed with fresh mealybugs. The observations about the mortality were recorded at 1 hr, 2 hr, 3 hr, 4 hr, 5 hr, 6 hr and 24 hrs of exposure to the insecticides.

6.4. Results

6.4.1. Residual toxicity of insecticides against female and crawlers of *P. solenopsis*

Results of film residual toxicity of insecticides applied at field dose to females of *P. solenopsis* are given in Tab. 6.1. Findings indicated that, both chlorpyrifos and malathion showed high toxicity (100 %) mortality to female mealybug at 24 hrs of exposure followed with dichlorvos (95.54 %), monocrotophos (82.84 %), alphamethrin (58.34 %) and endosulfan (35 %). The china leaves which were offered for feeding to *P. solenopsis*, had phyto-toxicity when treated with dichlorvos (Plate 6.2).

In case of residual toxicity to crawlers of *P. solenopsis* (Tab. 6.2) results showed that, all the insecticides registered 100 per cent mortality except endosulfan (25 %) at 1 hr of exposure.

6.4.2. Residual toxicity of insecticides against the predators of *P. solenopsis*

Results indicated that residual toxicity of all the insecticides was very high and caused 100 per cent mortality to the adults of *C. septempunctata* as against lowest in endosulfan (53.33 %) at 24 hrs (Tab. 6.3). Similar trend was observed for the grubs of *C. septempunctata* (Tab. 6.4) wherein treatments did not statistically differ significantly among each other. With regards to residual toxicity of insecticides to the adults of *C. sexmaculata* all the insecticides registered 100 per cent mortality at 24 hrs of exposure as compared to endosulfan (66.67 %). Based on mean mortality endosulfan was lesser toxic (Tab. 6.5). Residual toxicity of all the insecticides was extremely high registering 100 per cent mortality at 24 hrs of exposure to the grubs of *C. sexmaculata*, adults and grubs of *N. regularis*, adults and grubs of *S. coccivora*, adults and grubs of *H. maindroni*. There was no significant
difference among the insecticides tested against these predators (Tab. 6.6, 6.7, 6.8, 6.9, 6.10, 6.11 and 6.12).

Interestingly, chlorpyriphos registered zero per cent mortality to the grubs of *C. montrouzieri*, followed by endosulfan and dichlorvos (26.66 %), at 24 hrs of exposure period. Other insecticides namely monocrotophos and malathion registered 100 per cent mortality. Considering the mean mortality chlorpyriphos (21.66 %) followed by dichlorvos (34.16 %) were safer to the grubs of *Cryptolaemus* (Tab. 6.13). Chlorpyriphos treated grubs produced faint color adults due to chlorpyriphos toxicity (Plate 6.1). The insecticides *viz.*; chlorpyriphos, monocrotophos, malathion, dichlorvos and alphamethrin registered 100 per cent mortality to the grubs *Mallada* sp. at 24 hrs of exposure as compared to 46.66 per cent in endosulfan (Tab. 6.14).

6.4.3. Toxicity of insecticides incorporated honey (stomach poison) against the predators

With respect to stomach toxicity against the adults of *C. septempunctata* and *C. sexmaculata*, all the insecticides were very toxic and registered 100 per cent mortality at 24 hrs of exposure as against 60 to 70 per cent in endosulfan (Tab. 6.15 and Tab. 6.16).

6.4.4. Stomach toxicity of insecticides sprayed mealybugs against the predators

Result revealed that no mortality to the adults of *N. regularis* at 1 hr of exposure was noted due to treatment with chlorpyriphos and endosulfan. Besides, malathion caused very low (13.33 %) mortality as against 100 per cent by the dichlorvos (Tab. 6.17). Similar trend was noticed in case of *S. coccivora* (Tab. 6.18) and *H. maindroni* adults (Tab. 6.19).

6.5 Discussion

6.5.1. Residual toxicity of insecticides against the females and crawlers of *P. solenopsis*

Results indicate that except endosulfan and alphamethrin, the residual toxicity of all insecticides were significantly effective and caused higher mortality to the crawler and adult female mealybugs and in agreement with Morishita (2006) who reported that synthetic pyrethroids are less effective against Japanese mealybug, *Planococcus kraunhiae* than organophosphates. In the present study, endosulfan was
less toxic to *P. solenopsis* and is in conformity with Dhawan *et al.*, (2008). Similar observations with regards to chlorpyriphos and dichlorvos are in conformity with Suresh *et al.*, (2010). Phyto-toxic effects of dichlorvos observed on the China rose leaves in the laboratory may probably be due to fumigant action of the insecticide.

### 6.5.2. Residual toxicity of insecticides against the predators of *P. solenopsis*

Regarding residual toxicity of insecticides to adults and grubs of predators, results indicate that only endosulfan was safer to adults of *C. septempunctata* and *C. sexmaculata*. However, all the insecticides were very toxic (100 %) to grubs of *C. septempunctata*, *C. sexmaculata*, *N. regularis* and *S. coccivora* and *H. maindroni*. Endosulfan was found safer to the adults of *C. septempunctata* and *C. sexmaculata* due to its low residual toxicity than organophosphates. Makar and Jadhav (1981); Choudhary and Ghosh (1982); Babu (1988); Sharma *et al.* (1991) and Sonkar and Desai (1998) have already reported safety of endosulfan to the predatory coccinellid. In case of alphamethrin, grubs got knock down immediately after its exposure and remained moribund for quite sometime.

Interestingly, grubs of *Cryptolaemus* were selective to chlorpyriphos and dichlorvos and susceptible (100 %) to monocrotophos and malathion followed with alphamethrin (95 %). The studies are in agreement with Ramesh and Azam, (1987) who reported dichlorvos as safest and synthetic pyrethroids to be highly toxic to *C. montrouzieri*. It is inferred that the insecticides (Chlorpyriphos and dichlorvos) known for toxicity to coccinellids are safe to *Cryptolaemus*, which may probably be due to arrested activity of *Cryptolaemus* compared to other active coccinellids grubs. It was noted that other coccinellid grubs had profuse activity and thus imbibed lot of insecticides, leading to enhanced toxicity. Adults, which emerged from chlorpyriphos treated grubs were faint in color, and appeared a case of de-melanization. The behavioral resistance of *C. montrouzieri* against chlorpyriphos and dichlorvos affecting activity of spiracles is worth investigating by the Physiologists and Toxicologists.

With respect to *Mallada* sp. only endosulfan at field dose was found safer at various periods of intervals and are in agreement with Abida *et al.*, (2003) who reported residual toxicity of endosulfan as safer (mortality < 50 %) for 2nd instar grubs of Chrysopid, *Chrysoperla carnea*. 
6.5.3. Toxicity of insecticides-incorporated honey (stomach poison) against the predators

Result related to the stomach toxicity of insecticides incorporated honey revealed that only endosulfan was found safer to the adults of *C. septempunctata* and *C. sexmaculata*. As discussed above, the safety of endosulfan is again evidenced. So, it may be inferred that endosulfan is safer both as contact and stomach poison to the adult coccinellids.

6.5.4. Stomach toxicity of insecticides sprayed mealybugs against the predators

Result related to the stomach toxicity of insecticides treated mealybugs against the predators revealed that insecticides namely chlorpyriphos and endosulfan were safer while dichlorvos, most toxic. Present studies indicated the mean mortality range from 38.09 to 56.66 per cent for chlorpyriphos against the *N. regularis*, *S. coccivora* and *H. maindroni*, which is in agreement with Pasqualini and Civolani (2003) who evaluated the effects of 6 insecticides on adults of the coccinellids, *Adalia bipunctata*, *C. septempunctata* and *Oenopia conglobata* in fruit orchards revealed that chlorpyriphos caused 40.2 and 63 per cent mortality in apples and peach, respectively.

6.6 Conclusion

The insecticides *viz.*, chlorpyriphos and dichlorvos were highly toxic to mealybugs as well as to the majority of coccinellid and chrysopid predators. Among the insecticide studied, chlorpyriphos was found safer to *C. montrouzieri* and semi-selective to *N. regularis*, *S. coccivora* and *H. maindroni*. Chlorpyriphos being also a fumigant in action probably was found selective and safer to *Cryptolaemus* and warrants further investigations. Besides, reasons, behind the de-melanization of adult *Cryptolaemus* emerged from chlorpyriphos treated grubs also need to ponder upon. It may be noted that release of *Cryptolaemus* coupled with chlorpyriphos is likely to be a better option for the management of mealybugs infesting several horticultural and other crop plants.
Safety evaluation of insecticides on adult *Aenasius bambawalei* Hayat (Hymenoptera: Encyrtidae), a solitary endo-parasitoid of the mealybug, *Phenacoccus solenopsis* Tinsley

7.1 Abstract and Keywords

Six insecticides *viz*; chlorpyrifos 20 EC (0.1 %), endosulfan 35 EC (0.14 %), monocrotophos 36 SL (0.08 %), malathion 50 EC (0.25 %), dichlorvos 76 EC (0.3 %), and alphamethrin 10 EC (0.02 %) were tested for their stomach toxicity against solitary endoparasitoid, *Aenasius bambawalei*. Studies revealed that at the recommended field dose of six insecticides *viz*; chlorpyrifos, endosulfan, monocrotophos, malathion, dichlorvos and alphamethrin under laboratory conditions, residual toxicity was extremely high registering 100 per cent mortality to mealybug parasitoid, *A. bambawalei* at 24 hrs of exposure. However, when the field dose was reduced by twenty times, alphamethrin (55.23 %) followed by monocrotophos (78.57 %) were less toxic as compared to other pesticides. It was noticed that endosulfan as contact poison was more toxic (85.23 %) than as stomach poison (10.88 %) and unlike alphamethrin; which showed 55.23 per cent mortality as contact poison while 65.30 per cent as stomach poison. Only insecticide, which did not record mortality at 1 hrs of exposure, was alphamethrin.

It may be noted that, insecticides are likely to be less hostile in field conditions. Hence, lesser toxic insecticide to the parasitoid, *A. bambawalei* with substantial mortality to the mealybug may be a better strategy so as to conserve the parasitoid in crop ecosystem. Any study to safeguard the perpetuation of the natural enemies with the researches on the formulation of selective insecticide in near future is indispensable.

**Key Words:** *Aenasius bambawalei*, insecticides, *Phenacoccus solenopsis*, safety evaluation
7.2 Introduction

The solenopsis mealybug, *Phenacoccus solenopsis* Tinsley was noticed in 2004 for the first time on *Parthenium hysterophorus* L. in and around Delhi, which subsequently spread to over 64 plant species belonging to 27 families (Gautam et al., 2007, 2008). It was reported from field crops such as cotton, okra, tomato, brinjal, chilli and ornamental plants such as China rose, particularly in nine cotton-growing states of India, Punjab, Haryana, Rajasthan, Gujarat, Madhya Pradesh, Maharashtra, Tamil Nadu, Andhra Pradesh and Karnataka (Dhawan et al., 2007; Tanwar et al., 2007; Jhala et al., 2008; Dhara Jothi et al., 2008 and Bhosle et al., 2009). Within no time, it became a threat to Indian Agriculture. A well-known fact about the mealybug is that these are very hard to control due to their high reproductive rate, conceal habitats and impervious waxy covering to insecticides. The Biological control in the form of classical or augmentative or fortuitous control may forms the solution for such difficult to control pests. Survey conducted at Pusa campus (IARI) and nearby areas in Delhi indicated the presence of an encyrtid, a solitary endo-parasitoid from weeds *Parthenium hysterophorus*, *Xanthium strumarium* and *Achyranthes aspera* which was naturally parasitizing *P. solenopsis* on these weeds ranging from 20-70 per cent (Tanwar et al., 2008). The phenomenon of fortuitous biological control was also reported due to presence of an encyrtid parasitoid (*Aenasius* sp.) on exotic *P. solenopsis*, which parasitized 70-80 per cent mealybugs (Gautam et al., 2009). The parasitoid was subsequently described as *Aenasius bambawalei* for the first time from India (Hayat, 2009). As the parasitoid was found to be an efficient natural mortality factor for the newly flared up species of the mealybug under Indian conditions (Gautam et al., 2009), it can be a potent weapon to be used under biological control programme and that is why needs to be conserved.

Crops from which it was reported *viz.* Cotton, Brinjal, Okra are the one which consumes more than 50 per cent of India’s chemical pesticides and might lead to the extinction of *A. bambawalei* from this crops. So, it was the need of hour to have information about insecticides recommended for mealybug management at the same time lesser toxic to *A. bambawalei* and that’s why the present studies of safety evaluation of insecticides were carried out against the parasitoid, *A. bambawalei*. 
7.3 Materials and Methods

Details of the insecticides used for safety evaluation on solitary endo-parasitoid, *A. bambawalei*, are given in Tab. 2.1. Studies were carried out in Biological Control Laboratory, IARI, New Delhi at 27 ± 2°C temperature and 60 ± 5% relative humidity (RH).

7.3.1 Residual toxicity of insecticides on the parasitoid *A. bambawalei*

Experiments of residual film were carried out using standard methods of the IOBC/WPRS working group (Hassan, 1977 and 1992). Six insecticides *viz:* chlorpyriphos (0.05 %), endosulfan (0.07 %), monocrotophos (0.04 %), malathion (0.125 %), dichlorvos (0.15 %), and alphamethrin (0.01 %) at field dose and chlorpyriphos (0.0025 %), endosulfan (0.0035 %), monocrotophos (0.002 %), malathion (0.005 %), dichlorvos (0.0075 %), and alphamethrin (0.00005 %) at concentration twenty times less than their field dose were tested for their residual toxicity against *A. bambawalei* a solitary endoparasitoid, of *P. solenopsis*. The insecticides were tested at concentration 20 times less than their recommended doses because parasitoids were very susceptible to the field dose and they died within hours of an exposure to residual film. Glass cylinders (10×4 cm size) were taken and its walls were smeared with 1 ml solution of respective insecticide in acetone (see section 2.2.7, in chapter 2). A batch of glass cylinders coated with acetone only was kept as control and the experiment was replicated thrice. A due care was taken to coat only lower 2/3rd portion of the glass cylinders leaving upper 1/3rd portion residue free. Then the glass cylinders were shade dried for half hour. After shade drying the glass cylinders, a batch of well fed twenty (24 hrs old) adult parasitoids were released in to the glass cylinders. A small drop of undiluted honey was also smeared on top 1/3rd residue free portion of vial as a food supplement for parasitoids. The mouth of the glass cylinders was secured with black muslin cloth and tied with the rubber band. The observation about the mortality for experiments related to field dose was taken after 24 hrs of exposure to insecticides.

In another set of experiment, the parasitoids were exposed to twenty times lesser dose than the recommended doses. Besides, live parasitoids were transferred to the fresh untreated glass cylinder fed with honey, after completion of 6 hrs. Observations on the mortality of adult parasitoids were recorded at 1 hr, 2 hr, 3 hr, 4 hr, 5 hr, 6 hr and 24 hrs of insecticidal exposure.
7.3.2 Stomach toxicity of insecticides incorporated honey against the adult parasitoid, *A. bambawalei*

Six insecticides *viz*; chlorpyriphos 20 EC (0.1 %), endosulfan 35 EC (0.14 %), monocrotophos 36 SL (0.08 %), malathion 50 EC (0.25 %), dichlorvos 76 EC (0.3 %), and alphamethrin 10 EC (0.02 %) were tested for their stomach toxicity against solitary endoparasitoid, *A. bambawalei*. For stomach toxicity stomach poisons were made out of respective insecticide and honey (see section 2.2.8, in chapter 2). Honey was used for the preparation of a stomach poison because *A. bambawalei* at its adult stage feeds on honeydew excreted by the mealybugs. Glass cylinders (10×4 cm size) were taken and a batch of fifteen well fed adult parasitoids (24 hrs old) was released in to the glass cylinders smeared with respective stomach poisons. Batch of glass cylinders coated with acetone honey were kept as control and the experiment was replicated thrice. A small drop of undiluted honey was also smeared on top 1/3rd residue free portion of vial as a food supplement for parasitoids, so that they do not die of starvation. The mouth of the glass cylinder was secured with black muslin cloth and tied with the rubber band. The data was recorded at 1 hr, 2 hr, 3 hr, 4 hr, 5 hr, 6 hr and 24 hrs of insecticidal exposure. After completion of 6 hrs, live parasitoids were transferred to the fresh untreated glass cylinders fed with honey and final mortality was counted at 24 hrs.

7.4 Results

7.4.1 Residual toxicity of insecticides on the parasitoid, *A. bambawalei*

Results of residual toxicity of insecticides applied at field dose to parasitoid, *A. bambawalei* are given in Tab. 7.1. Studies revealed that at 24 hrs of exposure to field dose of the insecticides showed 100 per cent mortality. All the insecticides were significantly better than control but not among them.

Results of residual toxicity of insecticides applied at twenty times safer dose than field dose to parasitoid, *A. bambawalei* are given in Tab. 7.2. Out of six insecticides, chlorpyriphos and dichlorvos showed 100 per cent mortality at 1 hr of exposure as compared to alphamethrin (0.00 %) followed with monocrotophos (3.33 %). Considering the mean mortality irrespective of exposure period of insecticides, chlorpyriphos and dichlorvos were the most toxic (100 %) followed with monocrotophos (78.57 %) and alphamethrin (55.23 %).
7.4.2 Stomach toxicity of insecticides incorporated honey against the parasitoid *A. bambawalei*

Results of stomach toxicity of insecticide-incorporated honey against the parasitoid, *A. bambawalei* are given in Tab. 7.3. Studies indicate that per cent mortality at 1 hrs was lowest (0.0 %) for endosulfan and alphamethrin followed by monocrotophos (4.76 %) and malathion (23.80 %); whereas dichlorvos and chlorpyriphos were most toxic (100 %) at 1 hrs of exposure. However, at 6 hrs of exposure only endosulfan was significantly least toxic (4.76 %) as against other insecticides. Considering mean mortality irrespective of periods of exposure, endosulfan (10.88 %) was significantly safer than other insecticidal treatments to parasitoid.

7.5 Discussion

7.5.1 Residual toxicity of insecticides on the parasitoid, *A. bambawalei*

Studies with the field dose showed 100 per cent mortality of *A. bambawalei* for all the insecticidal treatments. It may be inferred that none of the insecticide is safe to the *A. bambawalei*, an efficient parasitoid of *P. solenopsis*. In fact parasitoids died within hours of exposure to all the insecticides. Dichlorvos and chlorpyriphos killed all the parasitoids within half hour of exposure. Studies with sub-lethal dosages, less than that of field dose, to ascertain the relatively safety of insecticides to *A. bambawalei* was inconclusive due to fast mortality in all the treatments at 24 hrs of exposure. At this dose dichlorvos and chlorpyriphos gave 100 per cent mean mortality which may possibly be due to fumigant action of these besides residual toxicity and in agreement with the findings of Yigit *et al.*, (1992) who reported harmful effects of Chlorpyriphos to *Leptomastrix dactylopii*, a citrus mealybug parasitoid in the laboratory as well as Brunner *et al.*, (2001) parasitoid, *Colpoclypeus florus*. In the present studies organophosphates were more toxic (100 %) to the parasitoid, *A. bambawalei* as supported by Wakgari and Giliomee (2003) who reported the contact toxicity of nine organophosphates insecticides ranging from 98-100 per cent mortality of mealybug parasitoid, *Coccidoxenoides peregrinus* caused (Hymenoptera: Encyrtidae) in less than 6 hours of treatment. Only alphamethrin with mean mortality of 55.23 per cent and monocrotophos (78.57 %) were comparatively lesser toxic might be due to less residual toxicity of both the pesticides; adding to it
monocrotophos is known for its systemic action rather than contact. Present results are in agreement with Brunner et al., (2001) who showed at reduced rates topically applied pyrethroids were low in toxicity to *C. florus*.

### 7.5.2 Stomach toxicity of insecticides-incorporated honey against the parasitoid *A. bambawalei*

Result indicated that endosulfan as contact poison was more toxic (85.23 %) than as stomach poison (10.88 %) and unlike alphamethrin; which showed 55.23 per cent mortality as contact poison while 65.30 per cent as stomach poison. Only insecticide, which did not record mortality at 1 hrs of exposure, was alphamethrin. These results are in agreement with Mani and Krishnamoorthy (1991) who did not report mortality of parasitoid, *Anagyrus dactylopii* at 1 hr, 3 hrs and 6 hrs of exposure to all tested pyrethroids. Both, dichlorvos and chlorpyriphos gave 100 per cent mortality and proved to be the most toxic insecticides to the adults of *A. bambawalei* and supported by Nilima et al., (2007) who reported chlorpyriphos to be the most toxic pesticide to the four species of parasitoids *viz.*, *Aphytis melinus*, *Eretmocerus eremicus*, *Encarsia Formosa* and *Gonatocerus ashmeadi*.

### 7.6 Conclusion

Studies revealed that the field dosages of six insecticides *viz.*; chlorpyriphos, endosulfan, monocrotophos, malathion, dichlorvos and alphamethrin under laboratory conditions were very toxic to parasitoid, *A. bambawalei*, an efficient parasitoid of *P. solenopsis*. It may be noted that, insecticides are likely to be less hostile in field conditions. Hence, lesser toxic insecticide to the parasitoid, *A. bambawalei* with substantial mortality to the mealybug may be a better strategy so as to conserve the parasitoid in crop ecosystem. Any study to safeguard the perpetuation of the natural enemies with the researches on the formulation of selective insecticide in near future is indispensable.
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Weekly weather data for the year 2007 from 1/1/2007 to 31/12/2007 of Indian Agricultural Research Institute, Pusa, New Delhi

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ACKNOWLEDGEMENTS

It is my privilege to express deep sense of gratitude to Dr. R. D. Gautam, Professor, Division of Entomology, Indian Agricultural Research Institute, New Delhi and Chairman of my Advisory committee, for his sustained encouragement, constructive criticism and imparting his enormous knowledge not only during my course of investigation but also in the preparation of the manuscript.

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Last but not least, I also pay my obeisance to lord Shivaji for his blessings and constant source of motivation during my carrier.

I am grateful to the Indian Agricultural Research Institute, New Delhi, for providing needed facilities to complete the research work.

Date 25.06.2011
Place Pusa, New Delhi (Sachin Suresh Suroshe)
10. ABSTRACT

Eco-Toxicological Studies on the Solenopsis Mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Coccoidea: Pseudococcidae) and its Predators

There are about 2000 species of mealybug occurring on about 250 plants families throughout the world and in India, more than 100 species of pseudococcids are known to attack a variety of plant species. Solenopsis mealybug, *Phenacoccus solenopsis* Tinsley, a pest of ornaments and fruit trees worldwide is cryptic in nature. It has already been recorded from 154 plant species belonging to 53 families comprising 20 field and horticultural crops, 45 ornamentals, 64 weeds and 25 bush trees. *P. solenopsis* has recently emerged as a serious sucking pest of India, attacking important field crops such as cotton, okra, tomato, brinjal, chilli and ornamental plants such as *Hibiscus rosa-sinensis*, particularly in the cotton growing belts of Punjab, Haryana, Gujarat, Maharashtra and Andhra Pradesh. As this mealybug is thought to be an exotic one and might have got introduced recently, the knowledge about its biology, natural enemy complex and management tactics was scanty in Indian conditions. Against this backdrop, studies were taken up on *P. solenopsis*.

Regarding, studies on survey of natural enemies of *P. solenopsis*, total of 17 parasitoids, belonging to seven families were found associated with various stages of *P. solenopsis* collected from Delhi, Maharashtra and Andhra Pradesh during 2007 to 2010. An encyrtid, *Aenasius bambawalei*, was recorded as a main primary parasitoid of *P. solenopsis*, which served as primary host of all hyper-parasitoids. Among hyper-parasitoids, *Promusciea unfasciiventris* was predominant. Interestingly, *Aphanogmus* sp. and *Anastatus* sp. are recorded for the first time on *P. solenopsis*. Besides, a total of ten predators *viz.*; eight coccinellids and two chrysopids were observed on *P. solenopsis*. On a good part not a single hyper-parasitoid is gregarious but solitary; otherwise a successful fortuitous biological control of *P. solenopsis*, with *A. bambawalei*, would not be possible.
Studies on population dynamics revealed that, Brinjal (var. shyamala) planted on 31st July 2007 recorded highest (33.8 adults) *P. solenopsis*, in March 2008 and lowest (0.93 adults) in November 2007, mealybugs found positively correlated with temperature. The other variety of Brinjal (var. Vikram) planted on 14th July 2008 was found negatively correlated with rainfall. The parasitoid, *A. bambawalei*, was found negatively correlated with *P. solenopsis* and *P. unfasciativentris*. Highest (53.73 adults) population of *P. solenopsis* was observed during November followed by October (21.13 adults) on sorrel planted in 2007. Highest (91.7 adults) population of *P. solenopsis* was observed during July followed by June (54.5 adults) on sorrel planted in 2008. Mealybug found positively correlated with temperature in sorrel planted 10.07.2007 and 15.04.2008. Highest number of *P. unfasciativentris* were recorded in October 2008 (116 adults) on sorrel and observed negatively correlated with mealybugs. Okra planted 26.06.2008, recorded highest (11.08 adults) population of *P. solenopsis* during October. It was positively correlated with temperature. Highest population of *A. bambawalei* was observed during October (23 adults), and found positively correlated with temperature and *P. unfasciativentris* which was highest in November (26 adults). China rose recorded highest (51.82 adults) population of *P. solenopsis* in July 2008, while lowest (0.06 adults) in October 2008. Mealybug was positively correlated with temperature, relative humidity and rainfall.

Result about biology of *P. solenopsis* showed that males went through 4 nymphal instars as against the 3 in females. The total life cycle for female and male was noticed as 14.06 ± 1.09 & 15.53 ± 0.83 days, respectively. Pre-mating, pre-ovipositional, ovipositional and post ovipositional periods for females were recorded as 2.11 ± 1.26, 7.66 ± 0.70, 10.22 ± 2.68 and 3.77 ± 1.71 days, respectively. Females once mate did not mate again. Reproduction was sexual and ovoviviparous, wherein only mated females laid eggs; unmated females did not lay eggs. Studies showed that *P. solenopsis* females reproduced sexually and not parthenogenetically, so can easily be manage through biological means, because mealybugs reproducing parthenogenetically multiply exponentially during unfavorable conditions and surpass its natural enemies.
Studies, on effect of six recommended insecticides revealed that both chlorpyrifos and malathion showed highest toxicity in terms of mean mortality (100 %) to female mealybugs at 24 hrs of exposure as against lowest in endosulfan (35 %). Interestingly, chlorpyrifos and dichlorvos, which proved toxic to mealybug, were less toxic to the grubs of Cryptolaemus montrouzieri, registering only 21.66 per cent and 34.16 per cent mortality, respectively. These six insecticides when offered to the adults along with honey differed in toxicity as stomach poison at 24 hrs of exposure. Endosulfan registered mortality to the tune of 60 per cent and 70 per cent against Coccinella septempunctata and Chielomenes sexmaculata, respectively. However, stomach toxicity pertaining to the insecticide-sprayed mealybugs as prey offered to three viz; Nephus regularis, Scymnus coccivora and Hyperaspis maindroni indicated that the insecticides, chlorpyrifos and endosulfan were lesser toxic wherein mean mortality ranged from 38.09 to 56.66 and 50 to 65.71 per cent, respectively. Dichlorvos was the most toxic registering 100 per cent mortality for all 3 predators. Reasons behind the de-melanization of adult Cryptolaemus emerged from chlorpyrifos treated grubs need to ponder upon. It may be noted that release of Cryptolaemus coupled with chlorpyrifos and dichlorvos is likely to be a better option for the management of mealybugs infesting several horticultural and other crop plants.

Safety evaluation of field dose of insecticides viz.; chlorpyrifos, endosulfan, monocrotophos, malathion, dichlorvos and alphamethrin under laboratory conditions, revealed that residual toxicity was extremely high (100 % mortality) to mealybug parasitoid, A. bambawalei, at 24 hrs of exposure. However, when the field dose was reduced by twenty times, alphamethrin (55.23 %) followed by monocrotophos (78.57 %) were less toxic as compared to other pesticides. It was noticed that endosulfan as contact poison was more toxic (85.23 %) than as stomach poison (10.88 %) and unlike alphamethrin; which showed 55.23 per cent mortality as contact poison while 65.30 per cent as stomach poison. It may be noted that, lesser toxic insecticide to the parasitoid, A. bambawalei with substantial mortality to the mealybug may be a better strategy so as to conserve the parasitoid. So, studies on selective formulation of insecticides are indispensable to safeguard the natural enemies.
9. सारांश

भिलीबन, फिनेकोक्स सोलेनोप्सिस टिन्सले (हेमिपेटा: कोक्कोइडिया: स्यूडोकोक्सिडिया), तथा उसकी परभक्षी कीटों पर पारिस्थितिकी–विषात्कता का अध्ययन

विश्वभर में भिलीबन की लगभग 2000 प्रजातियाँ हैं जो लगभग 250 पादप कुलों पर पाई जाती है। भारत में स्यूडोकोक्सिडिया की 100 से अधिक प्रजातियाँ, अनेक प्रकार की पादप प्रजातियाँ पर आक्रमण करती हैं। सोलेनोप्सिस भिलीबन, फिनेकोक्स सोलेनोप्सिस टिन्सले जो विश्वभर में सजावटी एवं फलदायी वृक्षों का एक पीड़क है, गुप्त प्रकृति का होता है। यह पहले से ही 53 कुलों के अंतर्गत आने वाली 154 पादप प्रजातियों से रेकॉर्ड किया जा चुका है जिनमें 20 खेत एवं उद्यान संबंधी फसलें हैं, 45 सजावटी पौधे, 64 खर–पत्तवर एवं 25 झाड़ीनुमा बूस है।

हाल ही में पी. सोलेनोप्सिस भारत में एक गंभीर चुपके पीड़क के रूप में उभरा है जो महत्वपूर्ण कृषि फसलों यथा, कपास, भिष्णु, बैंगन, भिंति तथा सजावटी पौधों यथा, हिबिस्कस रोजा–साइबरेन्सिस पर आक्रमण करता है, विशेष रूप से झाड़ी, धतुराणा, गुजरात, महाराष्ट्र एवं अंध्र प्रदेश के कपास उत्पादक क्षेत्रों में इसका गंभीर प्रकोप देखा गया है। वृंक यह एक विदेशी भिलीबन समझा जाता है और हाल ही में बाहर से यहाँ आया है, इसके जीवविज्ञान, प्राकृतिक शून्य समूह एवं प्रबंधन उपायों के विषय में भारतीय परिस्थितियों में जानकारी लेना आवश्यक था।

इसलिए, पी. सोलेनोप्सिस पर यह अध्ययन किया गया।

पी. सोलेनोप्सिस के प्राकृतिक शून्यों के संबंध में संरक्षण अध्ययन में 2007 से 2010 के वर्षों में तिल्ली, महाराष्ट्र एवं आंध्रप्रदेश से कुल 17 परजीवीय एककित फिड जो सात कुल के अंतर्गत आते हैं। एक एक्सिटिंड, एलासियंस बाम्बेवालाइ, पी. सोलेनोप्सिस के मुख्य प्राथमिक परजीवीय के रूप में रेकॉर्ड किया गया जो सभी अति–परजीवीयों के लिए प्राथमिक अतिवेश का कार्य करता है।

अति–परजीवीयों में, प्रोम्युआइडिया एनफोसीयाटिव्स्ट्रिस प्रमुख था। एफेंक्सस प्रजाति एवं एनास्टेटस प्रजाति पहली बार पी. सोलेनोप्सिस पर रेकॉर्ड किया गया है। इसके अतिरिक्त, कुल दस परभक्षी यथा; आज कोक्सीनलिडोस्ट्री एवं दो क्राइसोपिदि, पी. सोलेनोप्सिस पर पाए गए हैं। एक अच्छी बात है कि कोई भी परजीवीय यूथी (समूह में रहने वाला) नहीं है और एकाधि रूप से मिलते हैं; अन्यथा ए. बाम्बेवालाइ से पी. सोलेनोप्सिस का सफल जैव–नियंत्रण संभव नहीं होता।

आबादी गतिकी से संबंधित अध्ययन दर्शाते हैं कि 31 जुलाई 2007 को बोए गए बैंगन (किस्म श्यामला) पर मार्च 2008 में पी. सोलेनोप्सिस की अधिकतम
संख्या (33.8 वर्ष) रेकार्ड की गई जबकि नवम्बर 2007 में न्यूज़लैंड संख्या (0.93 वर्ष) रेकार्ड की गई जो तापमान के साथ धनात्मक सहसंबंध दर्शाता है। बैंगन की 14 जुलाई 2008 में बोई गई एक अन्य क्रिम (विक्रम) का वर्ष के साथ ऋणात्मक सहसंबंध पाया गया। परजीवाभ ए. बामबेचालाई का पी. सोल्नोपिस से और पी. अनफेसियाटिवेंटिस के साथ ऋणात्मक सहसंबंध पाया गया। पी. सोल्नोपिस की अधिकतम आबादी (53.73 वर्ष) नवंबर माह के दौरान पाई गई, इसके बाद 2007 में बोई गई सोर्टिंग पर अक्टूबर में आबादी (21.13 वर्ष) का स्थान रहा। पी. सोल्नोपिस की अधिकतम आबादी (91.7 वर्ष), 2008 में बोई गई सोर्टिंग पर जुलाई माह के दौरान पाई गई और तापमान जुलू (54.5 वर्ष) का स्थान रहा। 10.07.2007 एवं 15.04.2008 में बुआई किए गए सोर्टिंग पर मीलिओं का तापमान के साथ धनात्मक सहसंबंध पाया गया। पी. अनफेसियाटिवेंटिस की अधिकतम आबादी (116 वर्ष) अक्टूबर 2008 में सोर्टिंग पर देखी गई और मीलिओं के साथ ऋणात्मक सहसंबंध देखा गया। 26.06.2008 को बोई गई उलटी पर पी. सोल्नोपिस की अधिकतम आबादी (11.08 वर्ष) अक्टूबर माह के दौरान देखी गई। इसका तापमान के साथ धनात्मक सहसंबंध था। ए. बामबेचालाई की अधिकतम जनसंख्या (23 वर्ष) अक्टूबर माह के दौरान देखी गई और इसका तापमान एवं पी. अनफेसियाटिवेंटिस के साथ धनात्मक सहसंबंध पाया गया जो नवंबर माह में अधिकतम (26 वर्ष) था। चालानारेज पर पी. सोल्नोपिस की अधिकतम आबादी (51.82 वर्ष) जुलाई 2008 में देखी गई जबकि न्यूज़लैंड आबादी (0.06 वर्ष) अक्टूबर 2008 में पाई गई। मीलिओं का तापमान, सापेक्षिक आर्द्रता एवं वर्ष के साथ धनात्मक सहसंबंध था।

पी. सोल्नोपिस के जीवविज्ञान संबंधी अवधारण के परिणाम दर्शते हैं कि नर में 4 जिम्मेदार अवस्थाएं होती हैं जबकि मादा में 3 होती है। मादा एवं नर का सम्पूर्ण जीवन-वक्र क्रमांक: 14.06±1.09 एवं 15.53±0.83 दिन देखा गया। मादाओं में संगम-पूर्व, अण्डनिक्षेपण-पूर्व, अण्डनिक्षेपण एवं अण्डनिक्षेपण के बाद की अवधियाँ क्रमांक: 2.11±1.26, 7.66±0.70, 10.22±2.68 एवं 3.77±1.71 दिन पाई गई। एक बार संगम होने पर मादाएं पुंज़ संगम नहीं करती। प्रजनन लैंगिक एवं अंडजागुरु या जिसमें केवल संगम करने वाली मादाएं अपेक्षशील देती हैं। अवधारण दर्शते हैं कि पी. सोल्नोपिस की मादाएं लैंगिक रूप से प्रजनन करती हैं। और इनमें अनिस्केजन नहीं होता। इस प्रकार से इस मीलिओं का जीवन विधियों से प्रबंधन किया जा सकता है। क्योंकि अनिस्केजन से प्रगुणित होने वाले मीलिओं प्रतिकूल परिस्थितियों में बहुत तेज़ी से आबादी बनाते हैं और इस प्रकार से अपने प्राकृतिक शुद्धताओं से बच जाते हैं।
इस भीमीवग के विश्वदृष्टि हर संस्कृत कीटनाशकों के साथ अध्ययन दर्शाता है कि क्लोरोपायरॉफॉस और मैलाथियॉन मादा मीलबग स के प्रति उपचार के 24 घंटे पश्चात अधिकतम विषाक्त थे जबकि एंडोस्टक्फे कम (35% मत्तता) था। क्लोरोपायरॉफॉस और हाइक्लोरॉफॉस जो मीलबग के विश्वदृष्टि विषाक्त सिद्ध हुए, क्रिस्टोलिस मोल्टिग्राइड के बाद के प्रति कम विषाक्त पाए गए और इनमें क्रमशः 21.66 प्रतिशत एवं 34.16 प्रतिशत मत्तता ही दर्शायी। इन छह कीटनाशियों को जब व्यास्त कीटों को श्याद के साथ खिलाया गया तो 24 घंटे पश्चात जहर के दृष्टि से इनकी विषाक्तता भिन्न-भिन्न थी। कोकसीनेला सेंटेमनडेटा एवं कालोमेंस सेंक्सेनूलेटा विश्वदृष्टि एंडोस्टक्फे द्वारा मत्तता क्रमशः 60 प्रतिशत एवं 70 प्रतिशत थी। तेनेफ्स रेफ्लेक्स, रिफेनस, कोकसीनेला एवं फ्लायफ्लेक्स मेनडोनाई पर कीटनाशियों के छिड़काव के परिणाम स्वरूप देखा गया कि क्लोरोपायरॉफॉस तथा एंडोस्टक्फे कम विषाक्त थे और उनके औसत मत्तता क्रमशः 38.09 से 56.66 तथा 50.0 से 65.71 प्रतिशत देखी गई। हाइक्लोरॉफॉस, लीनों परम्परागत हेये सराधिक विषाक्त (100% मत्तता) था। क्लोरोपायरॉफॉस के उपचारित बाद से निकलने वाले व्यास्त क्रिप्सोलिस के डी-मीलेनाइजेशन के पीछे कारणों पर विचार किया जाना चाहिए। अनेक उदाहरण संबंधी एवं अन्य फसलों को ग्रस्त करने वाले इन मीलबग के प्रवृंदनार्थ क्रिप्सोलिस को छोड़ने के साथ-साथ क्लोरोपायरॉफॉस का अनुप्रयोग, एक बेहतर विकल्प हो सकता है।

प्रयोगशाला अवस्थाओं में कीटनाशियों यथा, क्लोरोपायरॉफॉस, एंडोस्टक्फे, मोनोक्रोफॉस, मैलाथियॉन, हाइक्लोरॉफॉस एवं एल्फामेशन की खेत मात्रा का मीलबग परजीवाच, ए. वाज्बेव्यालाई पर सुस्त क्षुद्रान्त दर्शाता है कि, अवशेष विषाक्तता अत्याधिक (100% मत्तता) पायी गई। जब खेत मात्रा को 20 गुना कम कर दिया जाता है तो एल्फामेशन (55.23%) और उसके बाद मोनोक्रोफॉस (78.57%) अन्य कीटनाशियों की तुलना में कम विषाक्त पाए गए। यह देखा गया कि एंडोस्टक्फे, पेट्र-षिक की तुलना (10.88%) में, सम्पूर्ण-षिक के रूप में अधिक विषाक्त (85.23%) था जबकि एल्फामेशन ने सम्पूर्ण-षिक के रूप में 55.23 प्रतिशत मत्तता और पेट्र-षिक के रूप में 65.30 प्रतिशत मत्तता दर्शायी। यह लोट किया जा सकता है कि परजीवाच, ए. वाज्बेव्यालाई के प्रति एक कम विषाक्त कीटनाशियों को मीलबग के विश्वदृष्टि अत्याधिक विषाक्त राखेगा, परजीवाच के संक्रमणकार्य एक बेहतर रणनीति हो सकती है। इस प्रकार से मीलबग के प्राकृतिक शरुआत को सुरक्षित रखने के लिए कीटनाशियों की विशिष्ट फार्मूलेशनों का अध्ययन प्रमाणधारक है।
Solenopsis mealybug, *Phenacoccus solenopsis* has been reported from various localities of different ecological zones of the globe and has been the topic of research for insect taxonomists and applied entomologists in India due to its invasiveness, rapid spread, morphological and biological variations and the need for establishing an effective control strategy. During the last few years, mealybugs which were considered to be minor pests in many crops have acquired the status of major pests especially in cotton, vegetables and fruits. *P. solenopsis*, has recently emerged as a serious insect pest in India, attacking important field crops such as cotton, okra, tomato, brinjal, chilli and ornamental plants such as *Hibiscus rosasinensis*. Against this backdrop, in order to throw light on above said aspects thesis entitled “Eco-Toxicological Studies on the Solenopsis Mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Coccoidea: Pseudococcidae) and its Predators” was taken up in ‘Biological control laboratory, Division of Entomology, IARI, New Delhi’.

The study was focused on survey of natural enemies of *P. solenopsis* infesting different host plants, for advocating an efficient natural enemy for biological control programme. Bio-ecology and population dynamics of the mealybug was addressed to know the details about its life history and phenology of *P. solenopsis*, for utilizing the knowledge towards its management. Efficacy studies, of recommended insecticides against various stages of *P. solenopsis* and safety evaluation on natural enemies of *P. solenopsis* was also carried out to come with selective insecticide for natural enemies with substantial toxicity to the mealybugs.

### 8.1 Survey of natural enemy complex of *P. solenopsis*

Total of 17 parasitoids, belonging to seven families were found associated with various stages of *P. solenopsis*. The natural enemy complex comprising four primary parasitoids, eleven hyper-parasitoids as well as two unidentified chalcidoids were isolated from the samples. An encyrtid, *Aenasius bambawalet*, was recorded as a main primary parasitoid of *P. solenopsis*, which served as primary host of all hyper-parasitoids. Among hyper-parasitoids, *Promuscidea unfasciativentris* was predominant. Interestingly, *Aphanogmus* sp. and *Anastatus* sp. are recorded for the
first time on *P. solenopsis*. Besides, a total of ten predators viz.; eight coccinellids and two chrysopids were observed on *P. solenopsis*. On a good part not a single hyper-parasitoid is gregarious but solitary; otherwise a successful fortuitous biological control of *P. solenopsis*, with *A. bambawalei*, would not be possible.

### 8.2 Population dynamics of *P. solenopsis* and its natural enemies

Studies revealed that, Brinjal (var. shyamala) planted on 31st July 2007 recorded highest (33.8 adults) *P. solenopsis*, in March 2008 and lowest (0.93 adults) in November 2007, mealybugs found positively correlated with temperature. However, when planted 14th July 2008 remained free from the mealybugs except scanty population noticed during January 2009. The other variety of Brinjal (var. Vikram) planted on 14th July 2008 was found negatively correlated with rainfall (-0.21). The parasitoid, *A. bambawalei*, was found negatively correlated with *P. solenopsis* (-0.21) and *P. unfasciativentris* (-0.12). Highest (53.73 adults) population of *P. solenopsis* was observed during November followed by October (21.13 adults) and lowest observed in December (1.3 adults) on sorrel planted in 2007. Highest (91.7 adults) population of *P. solenopsis* was observed during July followed by June (54.5 adults) and lowest was seen in December (1.3 adults) on sorrel planted in 2008. Mealybug found positively correlated with temperature in sorrel planted 10.07.2007 and 15.04.2008. Highest number of *P. unfasciativentris* were recorded in October 2008 (116 adults) on sorrel and observed negatively correlated (-0.2) with mealybugs. Okra planted 26.06.2008, recorded highest (11.08 adults) population of *P. solenopsis* during October as compared to lowest (3.4 adults) in November. It was positively correlated with temperature. Highest population of *A. bambawalei* was observed during October (23 adults), and found positively correlated with temperature (0.68) and *P. unfasciativentris* (0.11) which was highest in highest in November (26 adults). China rose recorded highest (51.82 adults) population of *P. solenopsis* in July 2008, while lowest (0.06 adults) in October 2008. Mealybug was positively correlated with temperature (0.53), relative humidity (0.67) and rainfall (0.61). Mealybug, was again found positively correlated with parasitoid, *A. bambawalei* (0.70), hyper-parasitoid, *P. unfasciativentris* (0.97) and hyper-parasitoid, *A. purpureus* (0.73).
8.3 Biology of *P. solenopsis* on *Parthenium hysterophorus* in the laboratory

Result showed that males went through 4 nymphal instars viz., Ist (5.86 ±
0.82 days), IInd (5.5 ± 0.56 days), IIIrd (2.2 ± 0.41 days) and IVth (2.26 ± 0.45 days)
as against the 3 instars in females Ist (5.94 ± 0.82 days), II nd (3.70 ± 1.21 days) and
IIIrd (4.41 ± 0.50 days). The total life cycle for female and male was noticed as
14.06 ± 1.09 & 15.53 ± 0.83 days, respectively. Pre-mating, pre-ovipositional,
ovipositional and post ovipositional periods were recorded as 2.11 ± 1.26, 7.66 ±
0.70, 10.22 ± 2.68 and 3.77 ± 1.71 days, respectively. The adult longevity was
reported as 47.125 ± 7.01 for unmated females, 3.66 ± 0.81 for unmated males, 24.66
± 4.5 for mated females and 1.33 ± 0.50 days for mated males. Females once mate
did not mate again. Reproduction was sexual and ovoviviparous, wherein only mated
females laid eggs (167.77 ± 25.57/female); unmated females did not lay eggs. Life
span (Life cycle duration and Adult longevity) was noticed as 61.5 ± 6.95, 18.66 ±
0.82, 38.44 ± 4.95 and 17.13 ± 0.97 days for unmated females, unmated males,
mated females and mated males, respectively. Studies showed that *P. solenopsis*
females reproduced sexually and not parthenogenetically, so can easily be managed
through biological means, because mealybugs reproducing parthenogenetically
multiply exponentially during unfavourable conditions and surpass the population of
its parasitoids and predators.

8.4 Effect of recommended insecticides on the important predators and its prey
*P. solenopsis*.

Studies revealed that, both chlorpyriphos and malathion showed highest
toxicity in terms of mean mortality (100 %) to female mealybugs at 24 hrs of
exposure as against lowest in endosulfan (35 %). Interestingly, chlorpyriphos and
dichlorvos, which proved toxic to mealy bug were less toxic to the grubs of *C.
montrouzieri*, registering only 21.66 per cent and 34.16 per cent mortality,
respectively. These insecticides when offered to the adults along with honey differed
in toxicity as stomach poison at 24 hrs of exposure. Endosulfan registered mortality
to the tune of 60 per cent and 70 per cent against *C. septempunctata* and *C.
sexmaculata*, respectively. However, stomach toxicity pertaining to the insecticide-
sprayed mealybugs as prey offered to all the three species of predators indicated that
the insecticides, chlorpyriphos and endosulfan were lesser toxic to these wherein
mean mortality ranged from 38.09 to 56.66 and 50 to 65.71 per cent, respectively.
Dichlorvos was the most toxic registering 100 per cent mortality for all 3 predators viz; *N. regularis*, *S. coccivora* and *H. maindroni*.

Chlorpyriphos being also a fumigant in action probably was found selective and safer to *Cryptolaemus* and warrants further investigations. Besides, reasons, behind the de-melanization of adult *Cryptolaemus* emerged from chlorpyriphos treated grubs also need to ponder upon. It may be noted that release of *Cryptolaemus* coupled with chlorpyriphos is likely to be a better option for the management of mealybugs infesting several horticultural and other crop plants.

### 8.5 Safety evaluation of recommended insecticides on the solitary endoparasitoid, *A. bambawalei*, of *P. solenopsis*

Six insecticides viz; chlorpyriphos 20 EC (0.1 %), endosulfan 35 EC (0.14 %), monocrotophos 36 SL (0.08 %), malathion 50 EC (0.25 %), dichlorvos 76 EC (0.3 %), and alphamethrin 10 EC (0.02 %) were tested for their stomach toxicity against solitary endoparasitoid, *Aenasius bambawalei*. Studies revealed that at the recommended field dose of six insecticides viz.; chlorpyriphos, endosulfan, monocrotophos, malathion, dichlorvos and alphamethrin under laboratory conditions, residual toxicity was extremely high registering 100 per cent mortality to mealybug parasitoid, *A. bambawalei* at 24 hrs of exposure. However, when the field dose was reduced by twenty times, alphamethrin (55.23 %) followed by monocrotophos (78.57 %) were less toxic as compared to other pesticides. It was noticed that endosulfan as contact poison was more toxic (85.23 %) than as stomach poison (10.88 %) and unlike alphamethrin; which showed 55.23 per cent mortality as contact poison while 65.30 per cent as stomach poison. Only insecticide, which did not record mortality at 1 hrs of exposure, was alphamethrin. It may be noted that, lesser toxic insecticide to the parasitoid, *A. bambawalei* with substantial mortality to the mealybug may be a better strategy so as to conserve the parasitoid in crop ecosystem. Any study to safeguard the perpetuation of the natural enemies with the researches on the formulation of selective insecticide in near future is indispensable.