

more when compared to middle (41%) and young (3%) farmers. Majority of the farmers 62.5% had 2 to 8 years experience in drip irrigation. The impact analysis indicated significant impact of drip irrigation on yield efficiency, income efficiency, water use efficiency, labour use efficiency and

input use efficiency. The constraint analysis revealed that majority (91%) of the farmers reported that clogging of emitters is the foremost problem in drip irrigation followed by rat biting (72%) and high investment despite subsidy (68%).

Field survey in Maharashtra



Field survey in Jalgon district of Maharashtra state



Field survey in Jalna district of Maharashtra state

3.9: New eco-compatible pest management strategies

3.9.1: Bollworms

Push-Pull strategy for management of pink bollworm Pectinophoragossypiella Saunders

The 'push-pull' approach is an ecological based

novel pest management tool that utilizes attractant and repellent (deterrent) components in combination. Fatty acids and their methyl esters identified in the previous year were quantified using 99.99% purity standards and evaluated for oviposition deterrent effect. Upon quantification oleic, linoleic and palmitic acid were identified as major compounds from the faecal pellet extract



using methanol. Five moth pairs with two days mating period followed by ten days egg laying was standardized for bioassay. Bioassays were carried out under laboratory conditions using individual compounds with 99.99% purity.

A significant difference was observed with increasing concentration of individual compound

in case of oleic and linoleic acid. The test of significance did not hold good for palmitic acid where all concentrations were at par with control. The mean number of eggs laid in each set of experiments with three different fatty acids has been depicted in Figure 3.9.1. Palmitic acid did not show any significant difference across all the concentrations.



Fig 3.9.1: Evaluation of fatty acids as oviposition deterrent for Pink bollworm

Note: Bars followed by same letters are not significant at P = 0.05 Tukey's HSD (honest significant difference)

GC-MS protocol for extraction of plant volatiles that may play a role as attractants was standardized. For identification of attractant volatiles, squares and bolls from all the four cultivated species of cotton were directly collected in five different solvents. Acetone and methanol were found to be the best solvents for extraction of volatiles. The major compounds identified were α/β pinene, carene, γ terpinene, α copaene, caryophyllene and humulene.

Identification of oviposition deterrent for management of Cotton Bollworm Helicoverpa armigera

Semiochemicals are acceptable alternative for the management of insect pests as they alter the behavior of insects. Among the semiochemicals, oviposition deterrents are the most explored.

In previous year volatiles were identified from the feacal pellets of *H. armigera*. These compounds were quantified using the pure compound. The

quantification showed the presence of four major fatty acids Viz., linoleic acid (9,12-octadecadienoic acid), palmitic acid (hexadecanoic acid), myristic (Tetradecanoic acid) and stearic acid (Octadecanoic acid). The methyl ester derivatives were quantitatively negligible.

Two male and two female pairing was found optimum for conducting bioassay. Muslin cloth treated with desired concentration of identified compound was used as oviposition substrate with suitable control. Five different concentrations (0.2%, 0.4%, 0.6%, 0.8% and 1.0%) of each compound was tested.

The average number of eggs laid across all replications in each set of experiments with four different fatty acids are presented in Figure 3.9.2. The per cent deterrence in four major fatty acid treatments varied from 16.5-73.5% for palmitic, 27.3 - 68.2% for linoleic, 33.9 - 69.8% for myristic and 34.5 - 68.9% for stearic acid. Almost similar values were obtained when compared with methanol control.





Fig 3.9.2: Average number of eggs laid in different fatty acids

Novel dispensers to enhance the trapping efficacy of gossyplure in managing pink bollworm

Gossyplure, the pheromone compound of pink bollworm viz., was tested with new dispensers and trap designs in two separate field experiments during 2018-19 at Coimbatore. In the first field experiment, eight dispensers made of neoprene, polypropylene, polyvinylpyrrolidone, cellulose acetate, zeolite, silicone, paraffin wax and rubber were evaluated for their efficacy (Fig.3.9.3). The dispenser made of silicone (9.5 adult/trap/week) and polypropylene (9.1 adult/trap/week) were significantly superior to the standard rubber dispenser (7.3 adult/trap/week) in attracting the male moths of pink bollworm. The neoprene dispenser had highest catch (6.3 adult/trap/week) in first four weeks but had short persistence (7 weeks) compared to that of standard dispenser (13 weeks). Two successful dispensers (silicone and polypropylene) selected from the previous year's experiments were evaluated in larger plots of one acre under farmers' field in Solavampalayam village, Coimbatore and compared with the standard rubber dispenser. The polypropylene recorded the highest mean catch of 46.3 /week, while the silicon and rubber dispensers provided a weekly mean trap catch of 41.2 and 34.9 males, respectively.



Fig 3.9.3: Efficacy of different dispensers in mass trapping pink bollworm males on cotton (2018-19)

In the second field trial, eight different trap designs viz., wing trap, LED combo trap, funnel trap, delta sticky trap, bottle trap, PET trap, water trap and cone trap fitted with standard rubber dispenser were evaluated PBW (Fig. 3.9.4). The weekly mean catches of PBW adult



males were in the order of delta sticky trap, (13.15) >LED combo trap (12.69)>wing trap (11.39)>funnel trap (10.97) > bottle trap (5.67)>PET trap (4.21)>cone trap (3.64)> water trap (2.87).



Fig 3.9.4: Efficacy of different trap designs in mass trapping pink bollworm males on cotton (2018-19)

3.9.2: Sucking pests

Chemical cues mediating natural enemy and sucking pest interaction in cotton

Identification of volatiles emitting from the sucking pests that attract natural enemies have potential for field deployment to enhance natural enemies and helping in eco-friendly pest management practices.

Sucking pests (jassids and whitefly) were collected

from the cotton field and volatiles extracted with solvent methanol. 9-Octadecenoic acid esters were major compounds in both sucking pests followed by 9,12-Octadecadienoic acid (Z,Z)-, methyl ester, hexadecanoic acid, methyl ester and methyl stearate.

3.9.3: Diseases

Identification of cotton endophytes with potential biocontrol activity against major diseases

Fungal endophytes were isolated from *G. arboreum* and *G. hirsutum* and identified through morphological characters and ITS analysis. Among the endophytes *Curvularia sp.*(10), *Macrophomina sp.* (08) and *Alternaria sp.* (05) were found to be dominant. Antagonistic potential of endophytic fungi was tested against *Corynespora cassiicola*, Alternaria sp. and *Fusarium sp.* Among all the endophytes CEL 2 (*Nigrospora sp.*), CEL 5 and CEL 19 (*N. sphaerica*), CEL 38, CEL 41 (*Diaporthe longicolla*), CEL 48, CEL 55, *Fusarium sp.*, *Isaria sp.* and *Nigrospora sp.* were found promising in inhibiting growth of the test pathogens. Colony inhibition was highest in CEL 41 followed by CEL 48 against *C. cassiicola*.



Antagonistic potential of different fungal endophytes against Corynespora cassiicola under dual culture technique. Endophytes as, Row 1: 1-CEL 1, 2-CEL 2, 3-CEL 3, 4-CEL 4, 5-CEL 5; Row 2: 1-CEL 11, 2-CEL 12, 3-CEL 13, 4-CEL 15, 5-CEL 16 and Row 3: 1-CEL 17, 2-CEL 19, 3-CEL 20, 4-CEL 21, 5-CEL 22



Standardization of GC-MS protocol for identification of antimicrobial volatile organic compounds (VOCs)

GC-MS protocol for identification of antimicrobial VOCs was standardized using three different methods viz., Headspace based identification, Solvent based Extraction and Matrix based Extraction: Using these three methods, fungal endophyte CEL 19 (*N. sphaerica*) was tested for the production of VOCs. The total ion chromatogram of CEL 19 revealed a list of different VOCs wherein, four major compounds identified viz 1, 3 diethyl benzene, 1, 4 diethyl benzene, p-Cymen-7-ol and m-Ethylacetophenone were found to be antimicrobial.

Biological control potential of cotton PGPR mediated through mVOC's

Microbial Volatile Organic Compounds (mVOC's) produced by promising and well characterized species of Bacillus strains of PGPR's were Total 9 mVOC's were identified identified. including 3 benzenes, 2 ketones, one each from alcohol, aldehyde, naphthyl and phenol group compound. The analysis of mVOC's spectra indicated that relative abundance and type of compounds produced are strain specific. Benzene, 1,3-diethyl- and Benzene, 1,4-diethyl- were the major antifungal compounds (identity >95%) detected from cotton PGPR strains Bacillus cereus CICR-D3, Bacillus aryabhattai CICR-D5 and Bacillus tequilensis CICR-H3. Additionally, insect repellent compound Naphthalene and nematicidal compounds i.e. m-Ethylacetophenone and Ethanone,1-(4-ethylphenyl) were also detected. Under sealed plate bioassay test conditions, Bacillus cereus CICR-D3 imparted significant mycelial inhibition after 5 days followed by Bacillus aryabhattai CICR-D5 and Bacillus tequilensis CICR-H3 against fungal pathogen, M. phaseolina



Volatile bioassay co-inoculated with Bacillus cereus CICR-D3 strain and Macrophomina phaseolina

3.9.4: Nematodes

Induction of Systemic Acquired Resistance against phyto nematodes.

Bio formulations (Curcumin, neem oil, aspirin and cow urine in different combinations) were evaluated for induction of systemic acquired resistance against reniform nematode under field conditions. Effect of bio formulations on nematode population in field and cotton yield was evaluated. Best formulation observed was Aspirin + Curcumine followed by curcumin alone. Reduction in reniform nematode population in soil was up to 55% giving an increase of seed cotton yield by 4 q/ha over untreated control.

Use of Marigold species for management of root knot nematode

Three species of marigold, *Tagetes erecta*, *T.patula* and *T.tenuifolia* were evaluated against root knot nematode. T.patula was most effective in reducing galls by root knot nematode. Planting of marigold by one week before planting of host plant was found to be better than planting both Tagetes and host plant together. Influence of marigold as nematode repellent was most pronounced within diameter of 10 cm from the marigold plant.

Extraction and evaluation of bioactive metabolitcs produced by *Purpureocillium lilacinum*

Studies were conducted to extract and evaluate the



bioactive metabolites produced by a native isolate of nematophagous fungus, Purpureocillium lilacinum isolated from nematode suppressive soil in cotton ecosystem. Ethyl acetate extract of the culture filtrate (CF) caused maximum of 100% juvenile mortality and inhibition in egg hatching of Reniform nematode, Rotylenchulus reniformis under in vitro condition. GC-MS analysis of ethyl acetate fraction of CFC indicated the presence of 13 different compounds, some of which are known to have nematicidal, antimicrobial, cytotoxic and insecticidal properties. Actinomycin C2,2-Pentanone, 4-hydroxy-4-methyl- (CAS), Hexanoic acid, 2-ethyl-,oxybis(2,1-ethanediyloxy-2,1ethanediyl) ester (CAS), 2-benzyl-3,6-dioxo-5methylpiperazine, Nonacosanol (CAS), 1-Docosanol (CAS), 2,5-Piperazinedione, 3,6-bis(2methylpropyl) are known to have nematicidal/ nematostatic properties.

3.10: Bio-diversity of pests and natural enemies in cotton ecosystem

3.10.1: Bollworms

Population dynamics of Pink bollworm in North India

Monitoring of Pink bollworm (PBW) is regularly done in North Zone since 2012-13 from 5 districts (Faridkot and Bhatinda in Punjab; Sriganganagar in Rajasthan; Hisar and Sirsa in Haryana) for larval recovery at various stage of crop growth from different varieties. During 2018-19 in North zone, based on green boll destructive sampling no PBW larvae were recovered in Bollgard-II hybrids . In Non Bt varieties HS-6, Ganganagar Ageti and RS-2013 first incidence of bollworm was observed in the 35th SMW which ranged from 1.5 to 12.1, 1.96 to 10.3 and 1.1 to 7.3 respectively based on percent fruiting bodies damage. Larval recovery(%) ranged between 3.3-12.4 in 2018-19 at 140, 160 & 175 days after sowing.

Incidence of PBW on BG-II hybrids was recorded from village Palwan (Distt. Jind, Haryana). The incidence of PBW was confined around the Vardhaman Cotton Mills, Palwan. Infestation ranged from 10-100% in green and opened bolls and depending on distance from the cotton mill. Infestation was recorded only up to 1 km radius.

Monitoring of Pink bollworm field incidence in India

The per cent infestation of PBW in green bolls of BG-II at 90-100 days after sowing was observed in the cotton growing districts of Maharashtra Viz., Aurangabad (3.3%), Parbhani (2.2%), Buldhana (2.2%) while there was no incidence in Akola and Amravati. Infestation of BG II cotton in Gujarat was in range of 0 to 7.2% and was highest in Surendranagar district (7.2%). Similarly, infestation of PBW in BG-II cotton fields of South India (Telangana, Andhra Pradesh and Karnataka) was more or less same as that of central India.

Survey teams from ICAR-CICR, Nagpur visited 16 districts of Maharashtra (Yavatmal, Wardha, Nagpur, Chandrapur, Gadchiroli, Amravati, Akola, Buldana, Jalana, Jalgaon, Dhule, Nandurbar, Aurangabad, Nanded, Hingoli and Parbhani), 1 a district of Telangana (Adilabad) and 1 district of Madhya Pradesh (Chhindwada) during 15 Sept to 12 October 2018 (90-120DAS). The teams collected about 20 green bolls from randomly selected plants one acre area of each field. Total 315 locations from 60 talukas of 18 districts of 3 states were surveyed and 6502 green bolls were collected. The green bolls were dissected for presence of live PBW larvae in the laboratory. Out of 6502 green bolls, 114 bolls were found to be infested with PBW indicating overall 1.75% (Maharashtra-1.88%, Telangana-0.80% and Madhya Pradesh 0.45%) infestation. Out of 315 location surveyed, the infestation in only 23 locations were found to have crossed the ETL (\geq 10% infestation).

In the station trial at Nagpur, PBW infestation was negligible till September, however it started increasing with the progress of season. Both Bt and non Bt cotton were found to be infested by PBW however, higher infestation was recorded in the non-Bt cotton (Fig. 3.10.1).





Fig. 3.10.1: Pink bollworm infestation in non-Bt and Bt cotton during 2018-19 (Nagpur)

Genetic Diversity of pink bollworm: Sequence analys were done with good quality sequence of COI fragment 686 bp portion of COI region of 214 PBW populations from different regions of India (South-52, Central-129 & North-33) to determine the population genetic structure, distribution and genetic diversity in all three cotton-growing zones. Edited unique sequences were deposited in Gene Bank using Bankit (Accession numbers: MK652512-MK652704, MK775533-MK775550). Alignments of all 214 COI sequences revealed that 27 haplotypes are present in overall populations including 11 in north, 14 in central and 6 in south region. The H1 and H2 are most common haplotypes present respectively in 143 and 32 populations and hence these two haplotypes could be proposed as ancestral/ original haplotypes. The presence of a star-shaped haplotype network linked by one or two mutations together with the multiple common haplotypes support this hypothesis. In this study zone-wise clustering revealed that central zone recorded low level of Haplotype diversity (0.499) as compared to south (0.560) and north (0.595).

Parasitization of Pink bollworm:

Pink bollworm infested green bolls were collected from different cotton growing districts of India. Parasitization by *Apanteles angaleti* in Maharashtra was found in the range of 1.28 (Wardha) to 10 % (Nagpur). In Gujarat, highest parasitization was recorded in Junagadh population (62.5%) and lowest in Anand (7.3). In Telangana, Warangal location was highest parasitized (25.9%) followed by populations from Mahabubnagar (6.25%) and Nalgonda (4.65%). PBW larvae collected from Nagpur was also parasitized (33.33%) by *Bracon lefroyi*.



Bracon sp.on pink bollworm larva Larval parasitiods Bracon sp.

Natural infestation of larval parasitiod, *Bracon* sp. (Hymenoptera: Braconidae) at 3.8% was also recorded on PBW larvae at CICR Regional Station, Main farm, Coimbatore.

3.10.2: Sucking Pests

Population dynamics of sucking pests of Nagpur (Central India)

Jassid population peaked twice first in second fortnight of August and another in the first week of October. Thrip population was higher during second week of August till the month end followed by a decline. Natural enemies viz. spider and Coccinellids were few in number (**Fig. 3.10.2**)





Fig. 3.10.2: Population dynamics of cotton pests over the season in DCH 32

Significant variation was observed in jassid and thrips population among the genotypes. Significantly higher population of jassid (6.90 jassid/ 3leaves) and aphid (11.49 aphids/ 3 leaves) was recorded on Suvin and Suraj, respectively. Comparatively more thrips were found on Suraj (4.96 thrips/ 3 leaves) followed by RCH 2 (3.61 thrips/ 3 leaves). Populations of whitefly, mirid bugs and spiders were negligible during the crop season (Fig. 3.10.3).



Fig. 3.10.3 : Pest and natural enemies' population on five genotypes over the season

Population dynamics of sucking pest at Sirsa (North India)

In RCH-650 BG-II hybrid jassid population ranged from 0.2-5.5.5/3 leaves with peak population between 29th and 33rd SMW. Population of whitefly

was initially observed in 22^{nd} SMW (2.6 / 3 leaves) and peak activity occurred in 29^{th} SMW (31.2 / 3 leaves). Population of thrips ranged from 0-36.3 / 3 leaves which were first noted in 22^{th} SMW and peak activity was observed in 29^{th} SMW (Fig 3.10.4)

In HS-6, leafhopper population ranged fro 3 leaves and peak activity was observed SMW. Population of whitefly was i



observed in 22^{nd} SMW and peak activity occurred in 30^{th} SMW (36.4 / 3 leaves). Thrips population ranged from 0-41.5 / 3 leaves with peak activity in 25^{th} SMW

In Ganganagar Ageti, jassid population ranged between 0-5.8/3 leaves and peak activity was observed in 33^{th} SMW. Population of whitefly was initially observed in 22th SMW and peak activity was in 28^{th} SMW (28.1 / 3 leaves). Thrips

population ranged from 0-36.8 / 3 leaves and peak activity was observed in $29^{\rm th}\,SMW$

In RS-2013, jassid population ranged from 0-5.2 / 3 leaves and peak activity was observed in 33^{st} and 36^{th} SMW. Population of whitefly was initially observed in 22^{nd} SMW and peak activity occurred in 28^{th} SMW (29.5 / 3 leaves). Thrips population ranged from 0-26.6 / 3 leaves and peak activity was observed in 26^{th} SMW.



Fig. 3.10.4: Population dynamics of whitefly under unprotected conditions at Sirsa.

Within plant, distribution of whitefly indicated that the whitefly prefers to feed on lower canopy of the plant as compared to middle and upper canopy. Mean whitefly adults population/leaf recorded during the entire season of 2018-19 on upper, middle and lower strata was 13.8 (7.5-28.5) & 4.6 (3.3-5.6) respectively during different part of the day RCH650BG-II and 4.09 (2.5-6.1) & 2.8 (2.0-4.), 9.9 (6.08-20.25) & 4.97(2.82-8.13), 13.75 (6.7-30.2) & 4.4 (3.2-8.8) respectively on HS-6. Though whitefly prefer to lay eggs on the fresh leaves, the nymphal pattern indicated equal preference for fresh leaves located both in middle and lower strata as compared to upper strata leaves.

The life table analysis of whitefly starting from 34-44 SMW indicated maximum natural mortality of nymphal stage due to fungal/bacterial infection 13.4(8.0-26.1%) followed by dislodgement 12.5 (2.6-29.4%) followed by parasitization 10.1 (4.6-25.0%).

Species diversity of thrips

Samples were collected from one district of Haryana (Sirsa), 8 Districts of Maharashtra (Nagpur, Washim, Amravati, Akola, Buldhana, Jalgaon, Rahuri & Wardha) and 4 Districts Tamil Nadu (Coimbatore, Salem, Ariyalur & Perambalur) and documentation of important taxonomical characters is under progress.



3.10.3 : Diseases

Prevalance of Xanthomonas citri pv. malvacearum races of Cotton

Bacterial leaf Blight (BLB) caused by *Xanthomonas citri* pv. *malvacearum* is an important disease of cotton and cause considerable yield losses. Ten BLB infected cotton leaf samples were collected from different cotton growing areas of Maharashtra (6 no.), Gujarat (2 no.) and North India (2 no.) to study race diversity. Biochemical and molecular characterisation of collected isolates and marker assisted selection by validated marker CIR246 was carried out.



Leaves showing the Yellowish slimy bacterial typical symptoms of colonies of Xam on angular leaf spot (YDCA) mediun

Occurrence and distribution of *Tobacco Streak Virus* (TSV) in Cotton

TSV infestation in germplasm of Gossypium barbadense was estimated. Per cent disease incidence was maximum in SP (35.8 %) and ICB-71 (26.6%) with disease grade of 3. The maximum disease incidence was observed at 60 to 75 days after sowing. The symptoms of TSV infection were very distinct with necrotic spots dark purple in colour and also drying of squares. The per cent disease incidences observed at 55 DAS in Gossypium hirustum (RCH 659 BG-II was 35.2% and Suraj Bt was 31.7%. Maximum disease incidence of TSV was observed at 70 days after sowing (DAS) in Mallika BG-II with 47.2 % with higher thrip population. Infected leaves of CCB 29, ICB 25 and Suvin showing typical symptoms were used for sap transmission studies on cowpea seedlings (CO 7) (Fig.3.10.5). Symptoms like chlorotic lesions, necrotic lesions, necrotic spots, veinal necrosis, systemic symptoms, necrosis on petioles, stem necrosis and total necrosis were observed on cowpea seedlings at 3 to 7 days after inoculation. Viral inoculum was maintained artificially in different hosts under insect proof net house conditions (Fig.3.10.6). Necrotic lesions were observed in greengram, black gram and soybean.



Fig. 3.10.5 : Sap transmission studies on local lesion host-Cowpea CO7

(Artificial inoculation)



Fig. 3.10.6: Maintenance of virus inoculum (Artificial inoculation)

3.10.4 : Nematodes

Prevalence of plant parasitic nematodes in Vidarbha region of Maharashtra

Eleven districts of Vidarbha region of Maharashtra were sampled for prevalence of plant parasitic nematodes. Reniform nematode *Rotylenchulus reniformis* was the most prevalent nematode on cotton followed by *Hoplolaimus* and *Pratylenchus sp.* The prevalence has indicated that due to intensive cultivation and monocropping, the intensity of plant parasitic nematodes may increase in future. Based on damage threshold of *R. reniformis*, hot spot areas were identified where nematode damage may become evident in years to come.

3.11: Integrated Pest Management

3.11.1: Bollworms

Resistance monitoring in Pink bollworm

For monitoring resistance against Cry1Ac and Cry2Ab, PBW populations were collected from different locations across India. The resistance development of PBW on BGII and non-Bt cotton fields was monitored. In North India, 9 districts



from three states (Hisar, Fatehabad and Sirsa of Harvana, Mansa, Abohar, Bathinda and Faridkot of Punjab, Sriganganagar and Hanumangarh of Rajasthan), in Central India 24 districts from three states (Wardha, Yavatmal Washim, Hingoli, Nanded, Parbhani, Aurangabad, Buldana, Akola, Amravati, Rahuri, Jalgaon districts of Maharashtra; Khandwa and Pandhurna districts of Madhva Pradesh, Surat, Bharuch, Vadodara, Anand, Ahmedabad, Bhavnagar, Amreli Junagadh, Rajkot and Surendranagar districts of Gujarat), in South India 12 districts of four states (Guntur, Prakasham, Kurnool and Krishna in Andhra Pradesh; Karimnagar, Adilabad, Warangal and Khammam in Telangana, Srivelliputtur and Coimbatore in Tamil Nadu and Dharwad and Raichur in Kharnataka were surveyed.

The population from North India was susceptible to Bt toxins except population of Jind district of Haryana. Field infestation of PBW was nil across the season on Bt cotton in north India. Thirty five population of PBW was subjected to Cry1Ac and thirty two populations were subjected to Cry2Ab log dose probit assays. PEW populations from Parbhani, Bellari, Akola, Amreli, Khandwa, Aurangabad, Prakasham. Guntur, Nalgunda and Kurnool recorded 143, 148, 159, 191, 248, 250, 294, 295, 434 and 899 fold resistances to Cry1Ac over susceptible check. Akola, Amravati, Nalgonda, Bellari, Guntur, Yadgiri, Khandwa, Amreli, Surendranagar, Aurangabad, Jind, Rahuri, Jalgaon, Kurnool and Vadodara recorded 158, 230, 274, 385, 425, 428, 548, 560, 628, 638, 1070, 1350, 1411, 3074 and 3831 fold resistance to Cry2Ab over the susceptible check.

Evaluation of egg parasitoid *Trichogramma species* to control Pink bollworm

A field trial was conducted to evaluate egg parasitoid *Trichogramma bactrae*, *Trichogramma brasiliensis* and *Trichogramma chilonis* through inundative release in cotton. Two releases at flowering (40-55 DAS) and two releases at boll maturation (60-75 DAS) stage at weekly interval along with a botanical, a microbial and two insecticides at 60-70 DAS, 70-80 DAS and 80-90 DAS. The observation was taken at ten days interval from 100DAS.

On green bolls, observations were recorded on number of exit hole, number of mines on the epicarp, number of larvae and per cent locule damage at ten days interval for all the treatments. Most effective was cypermethrin 4%EC + profenophos 40% (T-7) with 12.24% locule damage. T.bactrae with 13.05 per cent locule damage was most effective among three tested *Trichogramma species*. Neem oil and Beauveria bassiana also were promising for controlling PBW (Fig. 3.11.1).



Fig. 3.11.1: Evaluation of egg parasitoid Trichogramma species to control pink bollworm Pectinophora gossypiella (Saunders) in field conditions



3.11.2: Sucking Pest

Insecticide resistance monitoring against Jassid

In order to determine the resistance of jassid to different insecticides in Maharashtra, jassid samples from four locations were subjected to bioassays using eight insecticides. The results indicated that, LC₅₀ of Flonicamid ranged from 0.009 mg/L (Nagpur) to 0.111 mg/L (Wardha), Monocrotophos 0.010 mg/L (Nagpur) to 0.045 mg/L (Wardha), Acephate 0.010 mg/L (Wardha) to 0.037 mg/L (Amravati), Imidacloprid 0.005 mg/L (Wardha) to 0.044 mg/L (Amravati.), Acetamiprid 0.015 mg/L (Wardha) to 0.039 mg/L (Nagpur), Thiamethoxam 0.0012 mg/L (Wardha) to 0.054 mg/L (Chandrapur), Spiromeifen 0.004 mg/L (Nagpur) to 0.048 mg/L (Wardha), and Clothianidin 0.009 mg/L (Wardha) to 0.038 mg/L (Amravati). Wardha populations were more susceptible to Flonicamid, Thiomethoxam, Acetamiprid, Imidachlopride, Acephate and Clothianidin.

Insecticide resistance monitoring in whitefly in North India

Organophosphate: The change in resistance ratio is affected not only by the insecticide use pattern in main crop (cotton) but on other alternate hosts also. Resistance ratio obtained in case of ethion (2.6-7.4, 1.8-11.0 & 3.9-5.1), Chlorpyriphos (3.6-11.3,7.7-10.5 & 3.9-6.0), Triazophos (28.9-43.2, 11.8-54.6 & 20.4-37.1) during 2016-17, 2017-18 & 2018-19 was attributed to the pesticide use pattern on cotton as well as on other alternate hosts crops. In case of monocrotophos it was 2.7-4.7 during 2018-19. The resistance ratio was comparatively less during 2018-19.

Neonicotinoid: The high resistance recorded in our study at CICR Regional Station, Sirsa for thiamethoxam (12.8-58.9,10.3-51.5 & 14.8-22.6 fold resistance ratio during 2016-17, 2017-18 & 2018-19) at different locations of the north zone low resistance Acetamiprid (1.0-7.3, 6.9-10.3 & 5.7-10.6) and Imidacloprid (1.03-2.6 during 2018-19) insecticide use pattern. Low resistance ratio was recorded in our study at CICR Dinotefuran (0.9-17.8, 1.5-9.8 & 1-4.2 during 2016-17, 2017-18 & 2018-19), and Clothianidin (0.78-1.07 & 0.72-0.98 during

2017-18 & 2018-19).

Synthetic Pyrethroid: As pyrethroid use is being discouraged. Low to moderate resistances to cypermethrin have been reported in the B. tabaci populations .Very low or minimal resistance for Bifenthirin (0.63-28.2, 22.7-24.1 & 0.62-28.2 during 2016-17, 2017-18 and 2018-19) followed by Fenpropathrin in the studies conducted by CICR is again a resultant of insecticide use pattern in cotton and other crops.

Insect Growth Regulator: Younger stages are generally more sensitive to this group of insecticides as compared to older stages. Among these insecticides Difenthiuron (70.7-163.3, 33.3-128 & 48-66.8 fold resistance ratio during 2016-17, 2017-18 & 2018-19) was found with highest resistance ratio followed by Flonicamid (1.27-2.8, 0.47-2.8 & 0.6-2.8) during the last three years.

Identification of resistant genetic sources with mechanism of resistance against Jassid

Field screening of 54 genotypes of G. Hirsutum for identification of resistance/ tolerance against cotton leafhopper indicated that 13 and 24 genotypes were found superior and numerically on par with resistant check respectively in terms of leafhopper population. Leaf anatomical characters namely distance of phloem from the lower epidermis, phloem length, length of the palisade cell, leaf thickness, trichome type, length and density, the parameters linked with resistance for the hoppers, were analysed using leaf section. In general branched trichomes have predominantly been observed over non branched trichomes in the tolerant genotypes. Very sparse trichome hairs were observed in the susceptible line DCH 32. In comparative anatomical measurements genotypes namely NDLH 2010, AKH 2012-8, RS 2711, H1454, H 1464 Pusa 5760, GISV 216 and F2164 were distinctly identified for jassid tolerance among all other genotypes (Fig. 3.11.3). No significant difference in the biochemical parameters (chlorophyll a, total chlorophyll, leaf moisture) between the genotypes and the resistant check under protected and unprotected conditions were recorded except free amino acid content in 12 genotypes were lesser than the resistant check.





Fig. 3.11.2: Comparison of leaf anatomical characters

Evaluation of different groups of insecticides against thrips in cotton

Efficacy of different group of insecticides (10), biopesticides (3) and essential oils (5) were evaluated against thrips at Coimbatore and at Sirsa. Among insecticides, Spinoteram (64% reduction over control) followed by Fipronil (62% reduction over control) found highest efficacy against thrips. Among biopesticides, *Metarrhizium anisopliae* and among essential oils, neem oil followed by castor oil recorded higher efficacy against thrips. Biopesticides and essential oils were found safe against natural enemies viz., coccinellids and spiders.



*Mean value of two sprays

Fig. 3.11.3: Efficacy of different insecticides against Thrips under field condition

Evaluation of different colour sticky traps against thrips in cotton

Six colour sticky traps (Yellow, Blue, Red, Orange, Green and White) were evaluated against thrips. Among them blue colour followed by yellow attracted more number of thrips.

Bioefficacy of insecticides and biorationals against thrips

Under the common trials conducted for GEAC approved genotypes and testing of agrochemicals

during 2018-19, the efficacy of 12 label claim insecticides and five biorational interventions (castor oil, pongamia oil, sesame oil, 2 neem based formulation) were tested under laboratory conditions against thrips at three dosages during 2017-18 & 2018-19. Among the insecticides Spinosad (78 & 80%), Fipronil (72 & 82 %), Spinoteram 11.7% SC (68 & 72.2%) and Diafenthiuron (66 & 61.1%) followed by Profenphos gave highest mortality. Whereas among the biorational approaches sesame oil (58 &



41.1%), castor oil (50 & 50.1%), pongamia oil (32 & 56.7%) were recorded with moderate mortality against thrips.

Under field conditions among the insecticides Spinosad (68.1% reduction), fipronil (71.3%), spinoteram 11.7% SC(62.0%) and diafenthiuron (53.3%) and profenphos (58.1%) gave highest reduction in thrips count. Whereas among the biorational approaches sesame oil (35.2%), castor oil (44.9%), pongamia oil (27.0%) were recorded with moderate mortality against thrips.

Comparative field study of bio-insecticides and chemical insecticides

The field trials conducted at Sirsa revealed that the bioformulation of entomopathogenic fungal strain and chemical treatments were significantly superior over control in terms of whitefly nymphal mortality. The highest nymphal mortality at Sirsa on the seventh day after spray was recorded with Beauveria bassiana -4511 followed by Isaria javanica CICR-RSS -0102 and Pyriproxyfen (2.5ml/L), while at Nagpur, the highest nymphal mortality at 10 DAI were recorded with Fusarium moniliformae CICR-RSS-083 followed by B. bassiana-4511, I. javanica -CICR-RSS -0102 and Pyriproxyfen (2.5 ml/L). These treatments were significantly superior to Diafenthiuron 50% WP (1 g/L), Neem oil (300 ppm) and commercial formulation of L. lecanii (0.1% WP) (Fig. 3.11.4).



Fig. 3.11.4: Effect of selected entomopathogenic fungal strains and pesticides on whitefly nymphal mortality under field conditions at CICR RS Sirsa (2017-18 and 2018-19)

Development of thermal tolerant strain of biocontrol agent, Acerophagus papayae for sustainable management of papaya mealybug, Paracoccus marginatus

Parasitization potential of two strains of *A. papayae* were evaluated at 35°C. Constant exposure of mealybug to high temperature severely affected the growth and development. However, the parasitoid could survive and develop in the second instar mealybugs at the same temperature. The Strain A was able to develop and emerge at 35°C. Evaluation of parasitization potential of high temperature selected *A. papayae* under net house conditions revealed that the strain A showed better efficiency in terms of parasitization and emergence. Mean percent parasitization and emergence was high in host plant papaya, the sex ratio of female to male ratio was also high in papaya and tapioca than in cotton.

Evaluation of selectivity of insecticides against different mealybug species and their major natural enemies associated with cotton, tomato, brinjal and papaya

The cultures of *Paracoccus marginatus*, *Phenacoccus solenopsis*, *Maconellicoccus hirsutus*, *Aenasius arizonensis*, *Cryptolaemus montrozerii*, *Acerophagus papayae* and *Chrysoperla zastrowii sillamii* were maintained under laboratory condition. Safety evaluation of 14 different insecticides against *Aenasius arizonensis* was studied by glass vial residue method. The results revealed that the insecticides *viz.*, Imidacloprid, Thiamethoxam, Profenofos, Acetamiprid, Cypermethrin, lambda cyhalothrin, Pyriproxyfen + Fenpropathrin, Clothionidin were found to be highly toxic to the parasitoids, whereas diafenthiuron and thiodicarb were less toxic.

Two mealybug species (*Phenacoccus solenopsis*, *Paracoccus marginatus*), and their predators *Chrysoperla zastrowii sillamii and Cryptolaemus montrouzieri* used for enzyme assay were exposed to different insecticide groups. Insecticide exposure influences the level of detoxifying enzymes present in the mealybug. The activities of detoxifying enzymes such as esterase (EST), mixed function oxidase (MFO) and glutathione s-



transferase (GST) were estimated in insecticide exposed mealybugs, P. solenopsis and P. marginatus. Esterase activity was significantly higher in P. solenopsis exposed to Diafenthiuron (9.22 µM napthol/min/mg protein) followed by Acetamiprid (8.487 µM napthol/min/mg protein) and the lowest activity was observed in P. solenopsis exposed to Profenofos (2.039 µM napthol/min/mg protein and Cypermethrin (2.744 µM napthol/min/mg protein). The activity of MFO was more in *P. solenopsis* exposed to Pyriproxifen + fenpropathrin(129.88 nM cyto/min/mg protein) combination products followed by Acetamiprid (67.40 nM cyto/min/mg protein). Highest activity of GST was recorded in Triazophos treated (0.0240 µmol/ml/min) P.solenopsis. In P.marginatus, significant increase in EST was observed in triazofos treatment (8.709 µM napthol/min/mg protein) followed by Imidacloiprid (5.782 uM napthol/min/mg protein). Increased MFO activity was noticed in Acetamiprid (99.47 nM cyto/min/mg protein) treated P.marginatus followed by Profenofos (85.28 nM cyto/min/mg protein). The specific activity of GST was significantly more in *P. marginatus* exposed to Cypermethrin (0.0178 µmol/ml/min) followed by Triazofos (0.0157 µmol/ml/min. However, all three target enzymes EST, MFO and GSTs were totally nil in *P. marginatus* exposed to pyriproxifen, flonicamid, pyriproxifen + fenpropathrin and diafenthiuron.

Effect of thermal stress on fitness traits of two mealybug pests, *Phenacoccus solenopsis* and *Paracoccus marginatus* and their parasitoids *Aenasius bambawalei* and *Acerophagus papayae*.

The developmental biology of mealybugs, *P. solenopsis*, *P. marginatus* under different temperature regimes *viz.*, 25, 28, 30, 32 and 35°C was studied. The temperature above 32°C hamper the development of *P. marginatus*. Increase in temperature reduced the duration of pre - oviposition and oviposition period and as well fecundity of individual mealybugs. Threshold of development from egg to adult decreased

gradually in both male and female insects. The threshold of development for egg and first instar was at par and it decreased in succeeding stages of development.

3.11.3: Diseases

Survey to identify the biotic and abiotic problems and cultural practices adopted by cotton growers

A field survey was conducted to understand the current cultivation practices being followed by cotton farmers and the biotic and abiotic problems being faced in North India especially sudden wilt and root rot. Survey data from 50 farmers of Rajasthan, Haryana and Punjab indicated that 94.6% of farmers have not done deep ploughing since last 5 years or more, 70% of farmers are following cotton-wheat-cotton cropping system, 48.6% of farmers have applied the first irrigation before 30 days as against the recommendation of 45-50 days after sowing. Among these, 35.1% farmers have followed all three practices- i.e. cotton-wheat-cotton cropping system, no deep ploughing and early irrigation. Twenty per cent of the farmers fields had the soil hard pan conditions, 11% have followed mustard-cotton-mustard cropping system, 5.4% fields were had high soil moisture and they were in paddy growing areas. Such fields also had the biotic problems 32.4% having root rot & nematode infection, 10.8% root rot, 8.1% root rot & termite and in 24.3% fungal foliar spots during the month of September onwards, while 29.7% cotton field were observed with the sudden wilting problem. Among the 29.7% cotton fields, sudden wilting problems during the month of September onwards were recorded associated with no deep ploughing, cotton-wheat-cotton cropping system and early irrigation (before 30 DAS), in last more than 5 years. The soil EC and pH in these fields ranged between 2.5 to 4.5 dS/m and 7.6 to 7.9 respectively. These conditions appeared to be the main reasons for parawil/sudden wilt followed by a biotic problems such as root rot, vulnerability nematodes, fungal foliar spots (71.4%), and termites/rootrot(28.6%).





Fig. 3.11.5: Predominant factors found to be responsible for wilting in cotton fields

Sudden wilt management in cotton during water logging condition

A field experiment was conducted at CICR RS Sirsa to evaluate different treatments for the management of sudden wilt in cotton during water logging situations. Two sprays of each treatment were applied at an interval of 24 hours with first spray immediately after symptom initiation (within 24 h). The observations were recorded seven days after the second spray. Highest wilt recovery was recorded with 50 ppm sodium benzoate (75%), followed by 10 ppm cobalt chloride (56.5%) (Fig. 3.11.6:).



Fig. 3.11.6: Sudden wilt recovery percent at 7 days post spray

Management of root rot in *G. arboreum* and *G.hirsutum* cotton

Field experiments was conducted during 2017-18 and 2018-19 to evaluate the comparative effect of seed treatment with biological, chemical fungicides and their combination on management of root rot under sick field conditions at CICR RS Sirsa. Twelve biological and chemical treatments were applied as seed treatment on two cultivars namely CICR-2 (*G. abrobeum*) and CSH-3129 (*G. hirsutum*) grown in root rot sick field. The pooled data of two years trials revealed that the combination of seed treatment with *Trichoderma harzianum* (10g/kg), *Pseudomonas fluorescens* (10g/kg) and mycorrhiza (20g/kg) gave the highest root rot reduction over control in both the cultivars (57.5 and 51%) upto 60 days after sowing. Next best treatments were seed treatment with *T. harzianum* and combined application of *T. harzianum* & *P. fluorescens* (Fig. 3.11.7).



Fig. 3.11.7: Effect of seed treatment with biocontrol agents and chemical fungicide on management of root rot under sick field experiment (Pooled data) at CICR RS Sirsa

3.12: Development of new Detection Methods, Tools and Protocols

Novel method for isolation of wax degrading bacteria

A novel method for isolation of wax degrading bacteria from mealybug was standardized. The media identified was equally effective in extraction of bacteria and it is quick and labour saving. No difference in virulence of bacteria isolated from this media was recorded.

Development of a simple and low-cost laboratory rearing technique for cotton pink bollworm

A simple and inexpensive method laboratory rearing of PBW was using freshly excised green bolls (~10 d old) of cotton (*Gossypium hirsutum* L.) was developed (Figure 1). The adult females of



PBW preferred to lay eggs on the bracts and sometimes also on the bolls rind, either singly or in a group of 2-3 eggs. The larvae fed and pupated within the bolls itself. The developing larvae can be removed at any stage of their development for morphometric studies, conducting bioassays, etc. The newly formed pupae can easily be removed and maintained till adult emergence. All the biological parameters of PBW reared by this method were reasonably comparable with literature reports on its rearing using different natural and artificial diets. The egg hatching and adult emergence were $89.9 \pm 3.6\%$ and $92.3 \pm 2.3\%$, respectively with a mean fecundity of 203.7 ± 38.8 per female and a mean generation time of 34.4 ± 0.6 days. The insect raised by this method retained its ability to infest field grown cotton. Up to five generations of PBW could be raised by this method. This method is easily applicable and less expensive, and it would be highly useful in understanding the impacts of climate change on *P. gossypiella* phenology mediated through alterations and or aberrations in nutritional status of its host crop i.e. cotton **Fig.3.12.1**.



Oviposition cage

Pupal rearing

Larva developing within boll

Fig. 3.12.1: Laboratory rearing of PBW on detached green bolls of cotton