



**Title: Emerging and key pests of Bt cotton- their characterization, taxonomy, genetic diversity and control.**

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**INTRODUCTION**

Bt cotton was cultivated over an area of 63 lakh hectares in 2007-08. Four events were currently commercialized and a total of 134 Bt hybrids were available to farmers in the year 2007-08. The insect pest and disease scenario is changing quite dramatically. Prior to the introduction of transgenics, bollworms were of serious concern. With the advent of Bt transgenics, bollworms receded, both, in importance and incidence. Sucking pests have increased in their numbers as well as in their damage potential leading to the increased use of chemicals for their management on Bt cotton. Whether insecticide use on Bt cotton has drastically reduced is a matter of contention- the spectrum of insects, pests and of molecules being used for their management has certainly changed. The focus of this project has been on incidence, damage losses caused & management of emerging pests on Bt cotton across the different centers in the country.

**OBJECTIVES**

1. To determine the occurrence, seasonal dynamics and assessment of damage potential of emerging and key pests.
2. To determine the taxonomic status and genetic diversity of emerging and key pests.
3. Assessment of avoidable losses due to emerging pests on Btcotton.
4. To determine cultivar association with emerging pests.
5. Evaluation of effective control methods (biopesticides, biological control and insecticides) and identification of eco-friendly management strategies.

**ACTIVITIES**

- Recording of observations on temporal occurrence,

damage intensity of emerging pests and entomophages under various cropping systems. Observations will also be recorded on crop, adjacent weeds, trees, shrubs and the interventions made by farmers.

- Data analysis to elucidate resurgence factors
- Insect specimens of emerging and key pests and pathogen specimens will be collected from farmer fields at all centers for conventional taxonomy and advanced molecular characterization.
- Insect specimens will be subjected to taxonomy
- 16SrRNA from pathogens and CO-I, CO-II regions will be amplified from insects. Fatty acid profiles will be examined from the specimens. The amplicons will be sequenced and subjected to phylogenetic analysis and virulence association.
- Field and laboratory experiments will be conducted to evaluate the most potent biological control, bio-pesticide and insecticide for eco-friendly management of emerging pests and pathogens

#### EXECUTIVE SUMMARY

Mealy bug and mirids are emerging pests of importance in the country on cotton. Avoidable losses due to mealy bug were 29-50% and by thrips 39-50%. Mirids cause yield losses of upto 25-30% in Nagpur while they cause yield losses upto 11% in Dharwad. Mirid bug in 3 districts Belgaum, Dharwad and Gulberga of Karnataka state was comprised of a species complex of three species *Creontiades bisecatense*, *Hyalopelpus lineifer* and *Campylommalivida*. The predominant mealybug occurring on cotton was *Phenacoccus solenopsis* (taxonomic tools) with minimal genetic diversity (molecular tools). *Cryptolaemus montrouzeri* was also mass multiplied on mealy bugs in the lab. A method for quantifying the degree of infestation of the mealy bug has been developed. Jassids were the key pest during October and were more in IPM plot than non IPM. Damage potential of Jassids was estimated to be beyond 65 DAS and of mirids 90 DAS. 30 isolates of grey mildew were isolated. Isolates from diploid cotton were more virulent than tetraploid and were morphologically different.

#### SALIENT FINDINGS

##### Occurrence, seasonal dynamics and assessment of damage potential of emerging and key pests.

Mealybug and mirids were seen as emerging insect pests though thrips, jassid and aphids continued to occur on cotton (data has been submitted in the AICCIP format by all centers. Of the two, mirids were seen in Nagpur and in Dharwad while the mealybug was present throughout the country, causing maximum damage in the North as compared to Central India.

Losses were minimal due to mealy bugs in South India.

#### 1. Taxonomic and genetic diversity in mealybug

Survey and taxonomic study conducted in 47 locations in the nine cotton-growing states of India to identify the composition of mealybugs occurring on cotton showed two species viz; the solenopsis mealy bug, *Phenacoccus solenopsis* (Tinsley), and the pink hibiscus mealybug, *Maconellicoccus hirsutus* (Green), *P. solenopsis* was found to be the predominant mealybug species that comprised 95% of the samples examined.

A method was standardized to multiply mealy bugs on sprouted potato. These mealy bugs do not multiply well on any other known natural sources used for mealybug multiplication. Procurement and multiplication of the naturalized predator, *Cryptolaemus montrouzeri* was carried out with mealy bugs multiplied in the lab at CICR, Nagpur.



Live samples of mealy bugs from 47 locations of cotton growing regions were collected in October- November. DNA was isolated from 700 healthy female mealy bugs from 47 locations in the F1 generation; PCR amplified using specific primers to amplify three nuclear genes. Primers corresponding to the 18s rDNA 1 alpha fragments of the O2 and O10 expansion regions of the large subunit ribosomal DNA were designed and used. PCR amplicons of approximately 350 bp and 700 bp were sequenced using double pass analysis in 140 samples representing 3-4 samples per location. All of them had the identical sequence except one of the three samples, from Sriganaganagar which could be a different species. The cotton mealy bug identified as *Phenacoccus solenopsis* (taxonomically) with narrow genetic diversity (molecular analysis) throughout the country led to the conclusion that the mealy bug is an exotic pest. Mitochondrial genome studies are also being carried out where in regions of the cytochrome oxidase I gene being sequenced for further confirmation.

#### 2. Species complex and description of Mirids:

Three species of mirid bugs were found attacking Bt cotton in Dharwad and Haveri districts out of which, a species of the brown mirid *Creontiades biseratense* (Distant) (*Miridae Hemiptera*) was found in large numbers (Fig.1). Adults of this



Fig. 1: Adult and nymph of *Creontiades biseratense* (Distant)

species are brownish in colour with dark brown T-shaped band on pronotum. Nymphs are greenish in colour with dark brown wing pads,

Another species of mirid bug which was noticed in these regions was *Hyalopeplus lineifer*(Walker) (Miridae Hemiptera). Major identifying character of this species is the presence of brownish parallel streaks on the pronotum. Distinct colour morphs have also been noticed in *Hyalopeplus lineifer* (Walker) (Fig, 2).

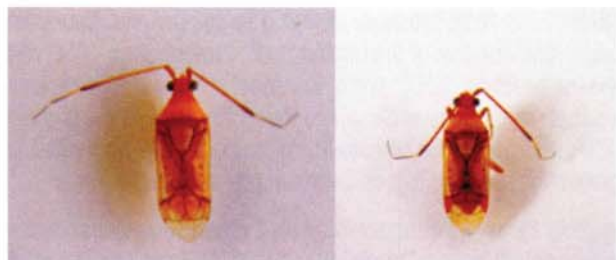


Fig. 2: *Hyalopeplus lineifer* (Walker) Black & Red colour morph

Nymphs of this species are creamish yellow in colour with long antenna and wing pad (Fig, 3),



Fig. 3: Nymph of *Hyalopeplus lineifer* (Walker)

Apart from these two, another species viz", *Campylomma livida* (Reuter) was noticed in large numbers on cotton. Mirids of this species are smaller in size with hairy wings.

### 3. Symptoms of damage due to Mirids :

Both adults and nymphs of mirids cause damage to cotton (Fig, 4 & 5), During feeding, mirids pierce the plant tissues with their



Fig. 4: Adult *Creonliades biseratense* (Distant) feeding on bolls



Fig. 5: Nymph of *Creonliades biseratense* (Distant) feeding on square

stylet. The affected area blackens that are seen as scars (Fig. 6), Feeding by these insects result in heavy shedding of squares and tiny bolls, In case of retained bolls, feeding results in the deformation, which is often referred to as "parrot beaking" (Fig. 7). If the infestation is severe in older bolls, the damaged locules may not develop properly and have stained lint (Fig. 8).



Fig. 6: Scars on bolls due to mired feeding



Fig.7: Parrot beaking symptom



Fig.8: Lint staining due to mired feeding

4. A method for quantifying the degree of infestation of the mealybug

The per cent infestation of mealy bug was recorded by counting randomly the number of plants infested by mealy bug. These infested plants were further analyzed to find out the severity of damage as per the following rating scale:

- 0 Nodamage
- I Scattered appearance of mealy bug
- II Fully infested on anyone branch of plant
- III Infestation on more than one branch/ half portion of the plant
- IV Heavy infestation in total plant

$$\% \text{ severity} = [\text{Average grade} / \text{Maximum grade}] \times 100$$

The method for quantification of mealy bug incidence and damage has now been fine-tuned and has been included as part of the AICCIP.

### 5. Documentation of host range of mealybug

Forty one alternate hosts were identified in Haryana including 10 weed hosts. During the off-season mealy bugs were found on eight weed hosts. Twenty two and five host plants were recorded from Punjab and Gujarat, respectively.

Avoidable losses due to emerging pests on 8t cotton

Yield loss due to mirids in Nagpur ranged from 25-30%. Damage potential of jassids was beyond 65 DAS and of mirids 90 DAS, Avoidable yield losses due to thrips in Gujarat on RCH 2Bt was 34,9-57,6 percent while for mealy bugs it was estimated to be 29.8- 50 percent,. The avoidable yield losses due to mirids (*Creantiades biseratense*) in Dharwad estimated on Bunny Btwas 11,69 percent.

The avoidable loss due to thrips were reported to be low in Punjab while mealy bug reported a very high damaging



potential inferred based on the yield obtained in treatments where mealy bugs were managed.

### Cultivar association with emerging pests

Association of emerging pests with Bt cultivars was not clear from experiments carried out in Coimbatore, Punjab, Nagpur and Haryana as a result of low pest incidence.

In Gujarat, KOCHH 441 Bt recorded the highest thrips number (17.9/ 3 leaves), MRC 7351 BG II recorded the highest whitefly population (8.9 adults/ 3 leaves) of the 64 genotypes, 58 Bt and 6 local checks tested.

In Madhya Pradesh, on the other hand, JKCH 99Bt, MECH 184, KOCHH-9632, Rasi Sai, Nandi Sandip 405 Akka BG I and Vishwanath recorded the maximum jassid nymphs per leaf (more than 5 nymphs/3 leaves) on par with one of the local checks RCH2 Non Bt in the year 2007-08.

It was reported that interspecific Bt hybrids (BG and BG II) record higher incidence of jassids, thrips and mirids as compared to intraspecific hybrids, in Oharwad. Mealy bugs did not show cultivar association.

Bacterial leaf blight was observed in all 36 Bt entries of Khandwa, with a POI of 11.88-27.92 percent. ACH-11-2BGII, NCS 207 Bt, RCH 2 Bt and Bembino recorded the highest incidence while Vishwanath Bt recorded the least along with the local check JK Hy-1. Myrothecium leaf blight recorded a POI ranging from 12-26.0 %. The highest POI was observed in Akka Bt followed by JK 4, Ankur 09 Bt and Alto Bt. Lowest POI was observed in Nandi Sandip.

### Effective control methods (biopesticides, biological control and insecticides) and identification of eco- friendly management strategies

None of the biologicals or botanicals included in this experiment, at the recommended doses, showed higher

efficacy against the target pests, especially jassids, over recommended doses of insecticides (Oharwad and Nagpur centres), in the field. In fact yield data revealed that these treatments were on par with untreated control while insecticides such as Imidacloprid, acephate and thiomethoxam out yielded control. On the other hand, insecticide treatments harbored lesser numbers of coccinellids over control and amongst these imidacloprid and thiomethoxam supported least coccinellid activity. This was true of the Oharwad centre that represents the irrigated zone while it was not observed in the rainfed region of Nagpur, that was representative of the rainfed zone. Profenophos 40EC and Monocrotophos 30EC were the most effective insecticides against mealy bugs at Sirsa and their efficacy was the highest 96h after spray. Oimethoate at the dose of 1.75 mill was the most effective against jassids and mealy bugs in Gujarat.

In a lab bioassay conducted against mealybug *Phenacoccus solenopsis* Tinsley, Chlorpyrifos 20EC and Profenophos 50EC resulted in cent per cent mortality of the *P. solenopsis* population followed by Triazophos 40EC (98.99%), Oimethoate 30EC (97.43 %), Methyl parathion 50EC (97.09%), Quinalphos 25EC (96.26%) and Acephate 75SP (96.26%). Thiodicarb 75 WP (95.05%), Acetamiprid 20 SP (86.06%), Thiomethoxam 25 WG (78.21 %) and Imidacloprid 17.80 SL (74.00%) showed better results. Biorationals Neem oil 0.03EC (77.13%) and Herbal product (72.00%) comparatively performed well, whereas *Verticillium lecani* (61.20%), *Beauveria bassiana* (55.02%), IGR Buprofezin 25 SC (64.32%) showed moderate mortality. Most of the effective organophosphates are extreme to moderately toxic according to WHO classification and are detrimental to several important natural enemies. Biorationals such as Neem oil, Herbal product, *Verticillium lecani*, *Beauveria bassiana*, Buprofezin and slightly hazardous WHO class: III organophosphate insecticide acephate can be part of *P. solenopsis* management strategy in light of their ecological safety.



Mirids (*Campylomma livida*)

