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Special Issue: Pink Bollworm

Photograph: Dr. Mahesh Upender, India

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Editorial

Pink bollworm can best be described as a mysterious pest. It sounds paradoxical, but the fact is that it is difficult to control but not difficult to manage. Difficult to control, because it is a cryptic pest that completes its larval cycle feeding inside a boll and is thus hidden away from insecticides and predatory insects. It is rather easy to manage, however, because the pest is specific to cotton and a cotton-free period of six months starves most of the pest populations.

Pink bollworm is also a mysterious pest because it is present in all the cotton growing countries of the world except Uzbekistan; it is a menacing problem in India and Pakistan but has been eradicated in the United States. The very fact that it was possible to eradicate the pest in such a huge continent as the United States strengthens the argument that it is not a difficult pest to manage, if not eradicate completely.

However, the PBW eradication story is a saga of collaboration, dedication, determination, persistence and scientific excellence. Government agencies in Mexico and the USA worked in close collaboration for 50 years, beginning in 1968, to eradicate the pink bollworm by 2018. The key components of the eradication programme were pest surveys using delta traps, sterile insect technique, pheromone-based mating confusion, short-season cotton and transgenic Bt cotton. Of all these, implementation of the 'sterile insect technique' strategy is the most amazing story. Three insect-rearing facilities were established in 1968, 1969 and 1995 in Texas and Arizona. The insect-rearing facility, measuring 6,131m^2 in Phoenix, Arizona, was able to produce an unbelievable 20 million to 28 million moths per day. The PBW eradication programme thus speaks volumes for the technical excellence, commitment and dedication of the USDA and US scientists, which will remain the 'gold standard' in the history of pest management — an achievement that is worth remembering and worth emulating.

Will India and Pakistan be able to manage the pink bollworm? Why are the two countries struggling to do it? The answer could be 'because the emphasis in both countries so far has been more on its control and less on its management'. Researchers in both countries have been looking at control measures using insecticides mostly, to be applied at economic threshold levels (ETLs) defined by pheromone trap catches. PBW management basically mandates the maintenance of a cotton crop-free window for at least six months. Maintaining a cotton-free closed-season for six months requires strict enforcement of a policy to ensure area-wide compliance of a fixed 10-days sowing window and a fixed termination date that does not allow any six-months old cotton crop to remain in the field, and also that all unharvested immature bolls are destroyed. Scientific reports confirm that most of the immature bolls harbour diapausing PBW larvae. Unfortunately, cotton in central and south India and in some regions of Pakistan is sown and harvested at staggered intervals to enable an area-wide window of cotton crop that stretches over eight to nine months every year thus providing a continuous source of food for the pink bollworm over a long period. Moreover, millions of diapausing pink bollworm larvae are safely harboured inside unharvested bolls mostly present in crop residues stacked or scattered near fields and in gin waste and trash near ginneries. Since a long season and diapausing larvae together cause the recurrent menace, it is logical that the two strategies, namely 'closed-season' and destruction of 'diapausing larvae', can have the greatest impact on PBW management.

Can these two strategies be implemented in India and Pakistan? The Indian hybrid Bt-cotton scenario provides ideal long-season conditions for the pink bollworm to eat well, survive, proliferate, develop resistance to Bt toxins and thus reclaim its lost status as the crown prince of cotton disasters. With the kind of crop conditions in India, PBW resistance to Bt cotton was inevitable; India grows hybrid Bt-hemizygous Bt-cotton on a near saturated scale to impose intensive selection pressure and non-Bt refuge is a mirage. Bt-hybrid seeds are expensive and are planted at a low density which forces farmers to maintain the crop through a long season to harvest a greater number of bolls from the fewer number of plants to get a decent harvest. As of now, timely termination of the crop to ensure a cotton-free window for six months on an area-wide scale appears to be nearly impossible in India but probably possible in Pakistan, because open pollinated varieties are grown there; seeds are inexpensive and high yields are possible within six months from a high-density crop. On the issue of destroying diapausing larvae, Pakistan scientists are seriously exploring harvesting technologies to destroy residual unharvested bolls (see Dr. Khalid Abdullah's interview in this issue), while Indian scientists are developing cotton gin trash treatment systems to destroy and prevent carryover of pink bollworm from ginning mills (see the article by Dr. Arude in this issue). Both technologies look promising.

The inspiration for this special issue on pink bollworm came from an online meeting of ICAC with cotton scientists of Pakistan on 8 February 2021. Pink bollworm occupied a major space in the discussions. It was clear that the scientific team was hungry for information on the pink bollworm and it is commendable that the cotton scientists in Multan appeared to be moving closer to robust management solutions. They are exploring several options including technologies for host plant resistance, mating confusion and removal of residual unharvested bolls.

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PBW is believed to have its origins in the Indo-Pak region from where it spread through seed-cotton to all cotton growing countries except in Uzbekistan. PBW is a serious problem in India and Pakistan, a seasonal problem in Egypt and Greece but not a problem in the USA and China. I invited short articles and interviewed scientists from all the countries mentioned above. Prof. Gutierrez and Prof. Tabashnik are legends in their own right and have expressed their views on the status of PBW in the United states and other countries. In their interview-responses, eminent cotton scientists, Dr Khalid Abdullah (Pakistan), Dr Mohamed Negm (Egypt), Dr YG Prasad (India), Dr GMV Prasada Rao (India), Dr Dong (China), Dr Yang (China) and Dr Stefanos (Greece), provide insightful inputs on the status and way forward for PBW management in their respective countries. There are 10 scientific articles on various aspects of PBW, authored by scientists from India, Egypt and Benin.

Indeed, with its lovely translucent pink shade, the pink bollworm is probably the most beautiful of all caterpillars in our agricultural ecosystems. I sincerely hope that this special issue will provide food for thought and ammunition to fight this beauty with our brains.

-Keshav R Kranthi

Photograph: Keshav Kranthi

India Must Return to IPM and Explore New *Bt* Genes

Interview with Prof. Bruce Tabashnik

Prof. Tabashnik is one of the most influential scientists of our times in entomology and biological sciences. He has led the Department of Entomology at University of Arizona for 24 years. His research team studies the evolution and management of insect resistance to insecticides and transgenic plants. Current work focuses on evolution of resistance to insecticidal proteins from the bacterium *Bacillus thuringiensis* (Bt). Tabashnik is Fellow of the Royal Society of Entomology, UK. He is the recipient of several awards which include the Nan-Yao Su Award for Innovation and Creativity in Entomology, Entomological Society of America; Koffler Prize in Research, Scholarship and Creative Activity, University of Arizona. He recently won the Plant-Insect Ecosystems Lifetime Achievement Award in Entomology from the Entomological Society of America

What in your opinion are the factors that could have been responsible for the delay of PBW resistance to *Bt*-cotton in China and the USA?

I think refuges of non-*Bt* cotton delayed PBW resistance to *Bt* cotton in China and the USA. *Bt* cotton planted in China produces one *Bt* toxin, Cry1Ac. In the Yangtze River Valley of China, when the percentage of all cotton hectares planted with non-*Bt* cotton dropped to 12% in 2008 and 2009, the proportion of pink bollworm larvae resistant to Cry1Ac increased significantly from 0% in 2007 to 2.6% in 2009. However, when the percentage of cotton planted with non-*Bt* cotton increased to 25% to 27% in 2011 to 2015, the proportion of resistant pink bollworm dropped back to 0%.

In the US state of Arizona, growers planted non-*Bt* cotton on at least 26% of their cotton hectares from 1997 to 2005 (mean = 38%). During that period, PBW remained susceptible to Cry1Ac.

What could have been the main reasons for the rapid development of PBW resistance to *Bt* cotton in India?

The scarcity of refuges of non-*Bt* cotton probably contributed to the rapid evolution of PBW resistance to *Bt* cotton in India.

PBW has become a serious menace in India in recent years on *Bt*-cotton. What could have been the reasons for resurgence in pestilence?

One of the main reasons for the resurgence of PBW in India is the pest's resistance to Cry1Ac, which is produced by the first type of *Bt* cotton, and to Cry2Ab, produced in the second type of *Bt* cotton in combination with Cry1Ac.

Which technologies held the key for PBW eradication in USA?

Bt cotton and releases of billions of sterile PBW moths by airplane over cotton fields were key for PBW eradication in the USA. Other tactics included application of PBW female sex pheromones and cultural controls such as restricting the growth period for cotton and destroying crop residues after harvest.

In your opinion, will implementation of strategies such as 1. 'refuge in bag', 2. 'sterile moth release' and 3. 'pheromone mating confusion' work for resistance management or pest management in India?

Because PBW resistance to Cry1Ac and Cry2Ab produced by *Bt* cotton is common and widespread in India, refuges of non-*Bt* cotton and other tactics are not likely to restore susceptibility to these toxins. Sterile moth releases and treatments with insecticides could be useful to reduce PBW populations.

Which strategies would you recommend for India for sustainable management or eradication of PBW?

For now, the widespread loss of the efficacy of *Bt* cotton against PBW in India means that a return to IPM emphasising other control tactics is essential. Recommended tactics for pink bollworm in India include planting of early to medium maturing cotton hybrids, termination of the crop by December, strict avoidance of ratoon cotton after harvest, removal or destruction of crop residues after harvest, deep summer ploughings, judicious use of insecticides based on scouting and thresholds, crop rotation, biological control with natural enemies, and pheromones for mass trapping and mating disruption.

I think eradication of PBW is not feasible without highly effective *Bt* cotton or another highly effective control that could be used in combination with the tactics listed above. It would be valuable to develop new *Bt* cotton that produces two or more toxins effective against PBW that are resistant to Cry1Ac and Cry2Ab. Candidates include Cry1B, Cry1C, and genetically modified Cry1A toxins.

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Photograph: Mahesh Upender

The Key to PBW Eradication Was Sterile Insect Technique Coupled With *Bt*-cotton

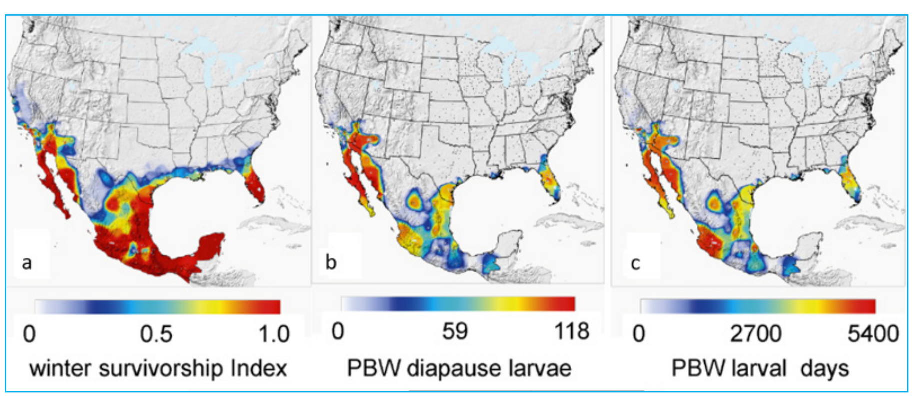
Interview with Prof. Andrew Paul Gutierrez

Prof. Andrew Paul Gutierrez FRES is Professor Emeritus in Ecosystem Science, College of Natural Resources, University of California at Berkeley, CA, USA. His research group investigates plant - herbivore-natural enemy interactions as driven by edaphic and weather factors using physiologically based tritrophic models. Prof Gutierrez is the CEO of CASAS Global NGO (http://www.casasglobal.org) which is dedicated to analyzing issues in diverse crops, rangelands, and medical and veterinary vectors to benefit populations and governments in developing countries worldwide. Prof. Gutierrez is the recipient of several Awards that include the Robert van den Bosch Medal and Member of IITA/Nigeria team receiving King Baudouin Award. Prof Gutierrez authored four books and more than 270 peer reviewed research papers. He guided 23 Ph.D. students, (1 MacArthur Fellow); 3 MS and 22 Postdoctoral fellows (World Food Prize)

What in your opinion are the factors that may have been responsible for the delay in PBW resistance to *Bt*-cotton in China and USA?

I do not know in detail the conditions in China; hence I will restrict my comments to the USA.

US cotton is mostly industrial scale, and when the exotic pink bollworm (native to South Asia) invaded the southwestern USA, insecticide use skyrocketed, secondary pest out breaks (bollworms, budworms, white fly, defoliators, plant bugs, etc.) became rampant, and yields and profit greatly declined. Agronomists soon developed short-season high-density cottons (SS-HD cotton) that were harvested and the stubble ploughed before PBW could produce overwintering, dormant larvae that would provide inoculum that infested the following season's crop. After a learning and development period with SS-HD cotton, high yields and reduced insecticide use returned, but these procedures required extra effort and costs. When *Bt* cotton became available in the late 1990s, open pollinated *Bt* cotton replaced SS-HD cotton because the costs were acceptable, and it was easier to implement, and pink bollworm proved highly susceptible to *Bt* toxins incorporated in various single and stacked configurations of cotton. However, because *Bt* toxins are insecticides, resistance was immediately recognised as a potential hazard to its sustainability. As noted by many authors on *Bt* resistance management in pink bollworm, the implementation and enforcement of the refuge strategy helped delay resistance development by maintaining a source of susceptible moths within the crop. The use of stacked *Bt* cottons further increased PBW susceptibility and made it more difficult for resistance to the different toxins to develop. In addition, the use of pheromone and sterile males SIT technologies reduced PBW populations regardless of their resistance status. The costs of these technologies were relatively small and government and state extension services oversaw their development and implementation. So it was a combination of all of these factors at different times that helped delay the development of resistance in PBW. Figure 1 shows the climatic limits of PBW in the USA.



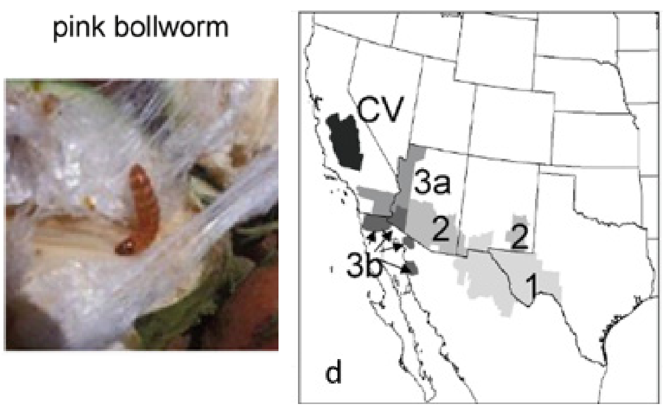


Figure 1. Geographic limits of pink bollworm in the southwestern USA: (a) cold weather limits as measured by winter survivorship indices, (b) the number of PBW larvae m^{-2} during the season, (c) cumulative larvae days during the season, and (d) the different phases of the PBW eradication program (see text, modified from Gutierrez and Ponti 2013).

What could have been the main reasons for the rapid development of PBW resistance to *Bt* cotton in India and Pakistan?

In contrast to the southwestern USA, cotton production in South Asia is mostly by millions of small-scale producers with most cotton being rainfed, and PBW is a native key pest having no effective natural controls. Prior to the introduction of *Bt* cotton to India beginning in 2002, insecticides to control the key pest PBW in long season cotton was increasing with expected outbreak of secondary pests (analogous native secondary pests such as 'American bollworm') leading to reduced yield, increased costs, and environmental issues. With the introduction of *Bt* cotton, there was initially an improvement, but predictably, resistance to insecticides and to *Bt* cotton began to arise. The major factors were the lack of consistent employment of refuges for resistance management due to scale and lack of extension infrastructure. Figure 2 shows how the refuge strategy would affect the time development of resistance in PBW. (Figure 2)

PBW has become a serious menace in India and Pakistan in recent years on *Bt*-cotton. What could have been the reasons for resurgence in pestilence?

In addition to the development of resistance to *Bt* toxins in PBW, insecticide use began to increase, and secondary pests not controlled by *Bt* toxin (white fly, plant bugs and *Bt* resistant defoliators, see Figure 3 below) increased, so that by 2012 insecticide use in India had reached pre-2002 levels, and Indian farmers were on both the insecticide and biotechnology treadmills. (Figure 3)

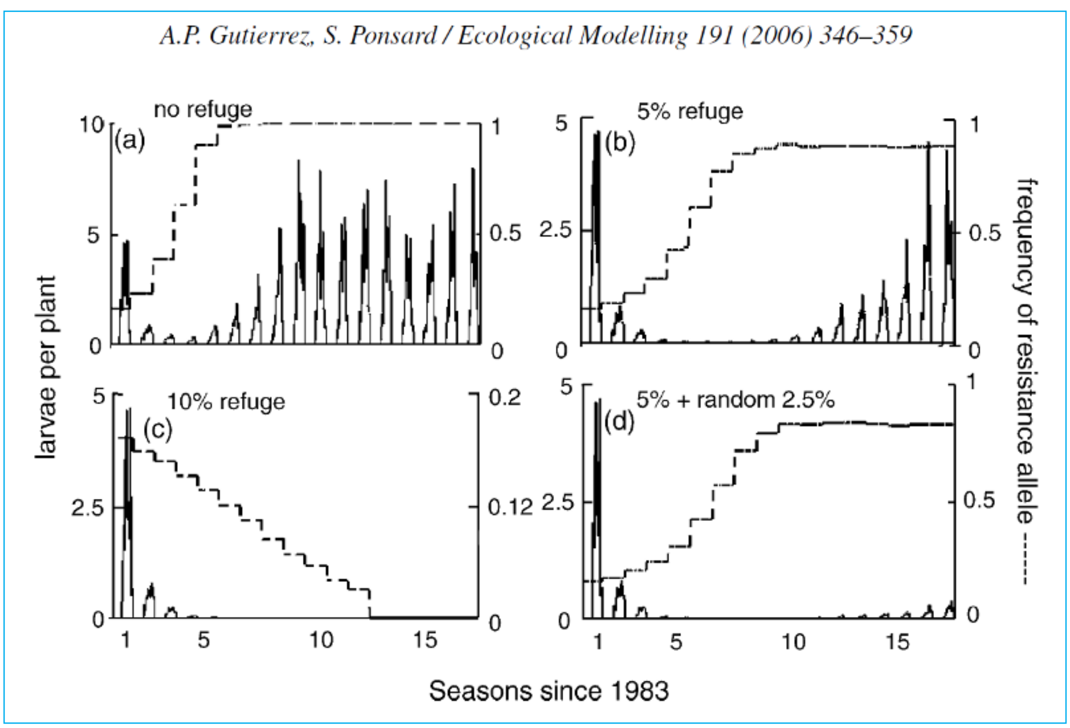


Figure 2. The potential development of resistance in PBW under different refuge conditions (Gutierrez, A. P and S. Ponsard, 2006. Physiologically based model of Bt cotton-pest interactions: I. Pink bollworm: resistance, refuges, and risk. Ecological Modelling 191:346-359).

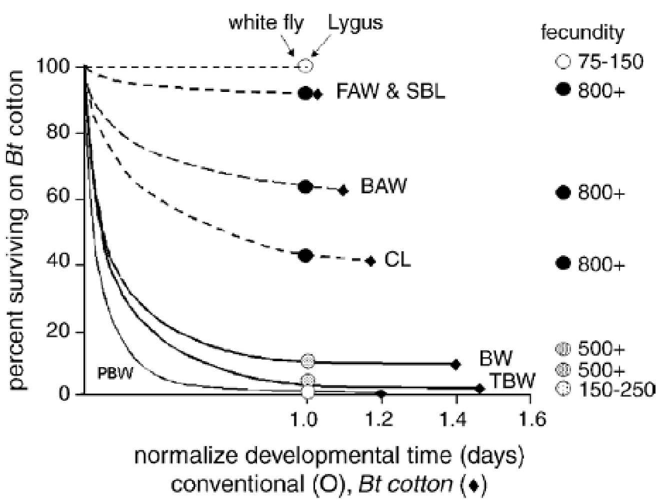


Fig. 3 The percent survival on normalized developmental time. Developmental time on conventional cotton is depicted by symbol shape ◯, while that of survivors on Bt cotton is indicated by (♦). Relative measures of fecundity are indicated on the right hand margin. Abbreviations are: whitefly (WF), *Lygus* bug (*Ly*), fall armyworm (FAW), soybean looper (SBL), beet armyworm (BAW), cabbage looper (CL), bollworm (BW), tobacco budworm (TBW), pink bollworm (PBW).

Figure 3, Relative susceptibility of various secondary pests in SW USA cotton (Gutierrez, A. P., J.J. Adamcyzk Jr. and S. Ponsard. 2006. A Physiologically based model of *Bt* cotton-pest interactions: II. bollworm-defoliator-natural enemy interactions. Ecological Modelling 191: 360-382.)

Which technologies held the key for PBW eradication in USA?

The key to PBW eradication was the suppression of PBW to very low levels by *Bt* cottons, that set the stage for effective use of sterile insect technique (SIT) on a regional level (see stages of the eradication program in **Fig. 1d** above). Without the suppressive effects of *Bt* cottons, SIT would not be effective as shown by SIT eradication attempts prior to the introduction *Bt* cotton.

In your opinion, will implementation of strategies such as 'refuge in a bag', 'sterile moth release' and 'pheromone mating confusion' work for resistance management or pest management in India and Pakistan?

The answer is no, because these technologies are expensive for poor farmers who largely lack the appropriate infrastructure to successfully implement them. The refuge in a bag approach of mixing non *Bt* seed with incredibly expensive hybrid *Bt* seed (unique to India) provides little net gain for farmers. The pheromone confusion approach didn't work in the USA and would be less likely to work in India, while SIT releases would need massive government intervention on a scale that dwarfs that attempted in the southwestern USA and Mexico, and worse the conditions for effective SIT are lacking (see map Fig.1)

Which strategies would you recommend for India and Pakistan for sustainable management or eradication of PBW?

What has been lacking are holistic methodologies for assessing the effects of weather on cotton/pest interactions – to enable rapid evaluation of scenarios. To see why in California, SS-HD cotton and post-harvest ploughing saved the irrigated cotton industry from the invasive pink bollworm by curtailing the development of dormant overwintering stages. The same methods in India show how rainfed SS-HD cotton would largely avoid infestation by adults emerging from winter dormancy in the various regions. Currently, the use of expensive effectively infertile hybrid *Bt* long season cottons limits planting densities and hence potential yields.

For India, field trial data show that pure-line short-season (SS), high-density (HD), non-GM rainfed varieties are a viable alternative to hybrid *Bt* varieties. Rainfed SS-HD cotton would largely escape pink bollworm infestation and the build up of late season pests. This would greatly reduce the need for insecticides, allow seed saving, and double yield and increase net income, and likely reduce suicides. (Figure 4)

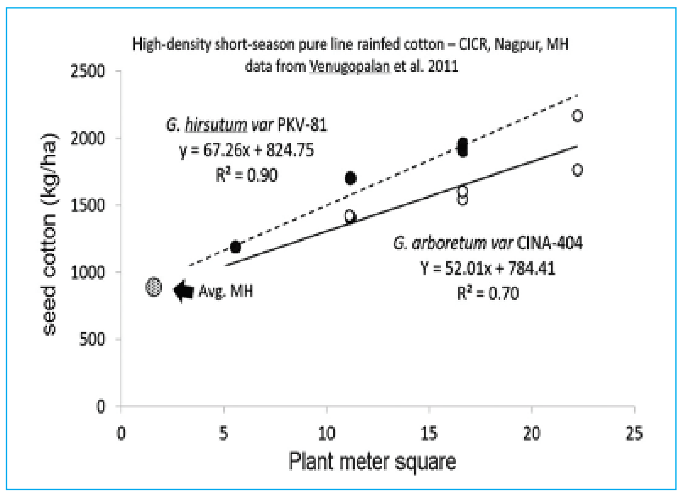


Figure 4. Seed cotton yields: average for Maharashtra for current hybrid *Bt* cotton (◉) and, field density trials of (●) *G. hirsutum* and (○) *G. arboretum* high-density short-season pure line cottons at CICR, Nagpur, India[13,15].

The hybrid *Bt* technology is incompatible with the SS-HD technology, because GM seed costs would greatly increase due to the higher seeding rates without commensurate increases in yield and would prevent seed saving. Cotton grown organically would increase profit, improve soils through reduced exposure to toxic chemicals and lower input costs, and reduce dependency on money lenders to cover productions costs. Fertile pure-line SS-HD non-GM cottons have been available for quite some time in India, but have not been widely implemented, and the obvious question is why? However, despite their utility, SS-HD cotton varieties are not a guarantee against the *gamble of the monsoon*, but they would lessen the impact.

Refuge Strategy and Resistance Monitoring are Critical for PBW Management

Interview with Dr Yuanxue Yang and Dr Hezhong Dong

Dr Yuanxue Yang is an assistant research fellow at Shandong Academy of Agricultural Sciences. Her research field is on cotton protection, focusing on pest management in cotton crop.

Dr Hezhong Dong is a senior principal scientist at Shandong Academy of Agricultural Sciences. His research field is on cotton ecology and physiology, focusing on field management for high yield and quality of cotton.

How serious is the problem of pink bollworm in different cotton growing regions of China?

Traditionally, there have been three major cotton growing regions in China, Yangtze River valley, Yellow River valley, and Northwest inland. Before *Bt* cotton was widely adopted in 2000 in China, the pink bollworm was one of the most serious insect pests of the nation, especially in Yangtze River valley. According to the estimates of cotton scientists and agronomists, it usually resulted in 15-20% and 5% yield reduction of cotton in Yangtze River valley and Yellow River valley cotton growing regions, respectively. However, Since the widespread of *Bt* cotton in 2000, the pink bollworm has not seriously occurred and is no longer a problem in China.

What could the range of economic damage (%) be in different regions? How serious was the pink bollworm in earlier times compared to what it is in recent years?

Because the occurrence varied with cotton growing regions, economic damage was also different in various regions. It was generally believed that the pink bollworm caused 15 and 5-10% economic loss in Yangtze River valley and Yellow River valley, respectively. However, the pink bollworm seldom occurred in the northwest inland, and thus there was no recorded economic damage in this region. It should be noted that the pink boll worm has not caused economic damage since wide adoption of *Bt* cotton in China.

What do you think are the factors that prompted its resurgence as a serious pest?

The pink bollworm currently is not a serious problem as the wide adoption of *Bt* cotton. However, if the pink bollworm produces resistance to *Bt* toxin, this factor will prompt its resurgence as a serious pest.

What in your opinion are the major factors that delayed its resistance development to *Bt* cotton in China?

China is one of the best countries to control the resistance to *Bt* cotton, which is largely due to the rich and diverse planting system and the use of F1 and F2 hybrid cotton seeds. There are many host plants of the cotton bollworm. The cotton-based intercropping system provides a natural refuge for the cotton bollworm. The host crop of pink bollworm is relatively single and other crops cannot provide refuge for it. However, hybrid cotton is widely planted in China. One of the parents is usually non-*Bt* cotton. The average purity of hybrid seed (F1) is 95-98%, which results in at least 3-5% of non-*Bt* cotton plants in the field and provides a refuge for pink bollworm. Therefore, intercropping, double cropping and hybrid seed delayed the resistance of boll worm or pink bollworm to *Bt* cotton.

Have there been any recent innovations for PBW management from research institutes in China?

In recent years, China's cotton planting has shifted to the Northwest. The cotton planting area in the Northwest Inland accounts for about 80% of the total national cotton area. However, pink bollworm rarely occurs in the Northwest inland and has not been a major pest, so the research on pink bollworm management is rarely carried out in recent years.

What in your opinion are the most important management strategies?

Refuge strategy and resistance monitoring should be the most important management strategies.

Photograph: Mahesh Upender

Pakistan is Exploring Triple Gene Transgenic Cotton, Destruction of Residual Bolls and Mill Waste and Mating Confusion Technologies

Interview with Dr Khalid Abdullah

Dr Khalid Abdullah is the Cotton Commissioner at the Ministry of National Food Security and Research, Government of Pakistan. Dr Abdullah is an eminent cotton scientist of international repute. Dr Abdullah spent 20 years of his career as active researcher, administrative and policy formulator. He authored a book, book chapter and wrote over 40 scientific papers, supervised 25 Doctoral and Post Graduate students in Entomology and Plant Protection. Dr Abdullah joined the Ministry in 2010. As Cotton Commissioner, he recommends policy guidelines and future strategies on cotton to the government. Dr. Abdullah has been actively involved in Technical Assistance Program for C-4 countries with Ministry of Commerce and Ministry of Textile Industry.

How serious is the problem of pink bollworm in Pakistan?

The pink bollworm (PBW) has been a serious pest since 2015 in Pakistan. It has been causing annual losses of 2-3 million bales in cotton production, either directly or indirectly. The graph above shows a small surge in population build-up in May-June, which is followed by a big peak of infestation and population build-up in August and September, that severely damages cotton crop. These two peaks have a different level of influence depending on the time of sowing. The recommended time of sowing in Pakistan is April May. Late sown cotton when planted in May-June becomes very vulnerable to the cotton leaf curl virus (CLCuV) leading to the crop being stunted in its early growth stages and bearing very few flowers thus resulting in poor yields. Therefore, wherever it is possible to advance sowing, especially in the absence of cotton-wheat rotation, farmers prefer early sowing or timely sowing of cotton crop to escape CLCuV. However, crops sown in March start flowering in May-June and support the first generation PBW larvae. The proportion of early sown crop is actually less. Usually less than 1.0% percent of the area is sown in first week of April 1-2% percent in second week of April and 2-3% percent in the 3rd week of April. Though less than 5-6% of the area is sown before mid-April, the early sown crop provides food for the early generation PBW and facilitates carryover of the pest into the later crop stages of the season. Most importantly, by the time, the timely sown cotton (after wheat) reaches flowering stage, the second-generation population of PBW would have sufficiently built up to damage early flowers and fruiting bodies. Farmers spray insecticides such as triazophos and its mixtures to control PBW because of which the problem gets worse, with whitefly resurgence as a consequence of insecticide applications. This triggers the beginning of an end, wherein cotton becomes unremunerative due to high cost of production, primarily through the additional cost of repeated insecticide applications, especially during the peak arrival stage of cotton. As the industrial procurement slows down and prices drop, farmers either abandon the crop or lose interest in investing more on crop management.

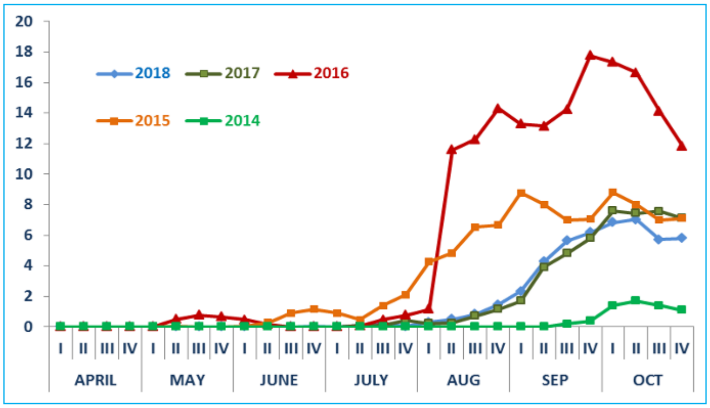


Figure 1. PBW hotspots (%) above ETL in Punjab
Source: Pest Warning and Quality Control Department, Punjab

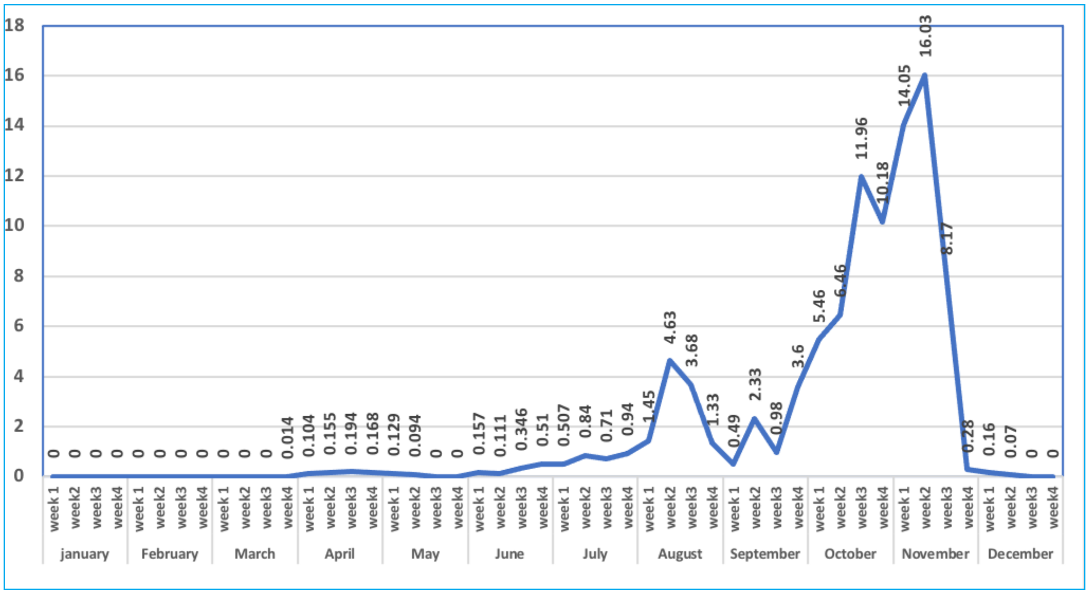


Figure 2. Month-wise PBW moth catches/trap at Multan during 2020.
Note that the trend is not very different from the previous years as shown in figure 1.

Another factor that contributes to PBW pestilence is the mill waste of ginning factories', where infested bolls are ginned and the trash along with larvae are thrown away. The mill waste becomes a strong source of infestation. Cotton stalks are usually stacked and stored near cotton fields and are commonly used as kitchen firewood in rural households. The infested immature un-picked bolls attached to cotton stalks also become a source of first-generation moths which emerge when temperatures get favourable and early sown cotton is available in fields.

What could the range of economic damage (%) be in different regions?

The direct production losses were estimated to be about 1-2 million bales in Punjab and 0.5-1 million bales in Sindh province, whereas the indirect losses could be much higher than that. Cotton production in Pakistan dropped to 9.18 million bales during 2019-20 which is expected to reduce further to less than 7.0 million bales in 2020-21. The damage due to rains is estimated be about 1.6 million in Sindh province during 2020-21. Cotton in Punjab was badly hit by PBW, whitefly and CLCV, while the crop in Sindh province suffered from a moderate PBW attack.

How serious was the pink bollworm in earlier times compared to what it is in recent years?

Pink bollworm was a serious pest in the past, during the pre-GMO period. However, the American bollworm *Helicoverpa armigera* which had developed resistance against major pesticide groups was a more serious pest than PBW. This doesn't mean that PBW was not causing damage prior to 2005. The Pakistan Central Cotton Committee (PCCC) introduced pheromone band as a male disruptive technique and found it to be very effective. PBW is no longer a serious pest in many advanced countries and hence the research backstopping has been far less than it was in yester years of the pre-GMO era.

What do you think are the factors that prompted its recent resurgence as a serious pest?

Several factors appear to have influenced PBW resurgence in Pakistan. After the patent expiry of BG-1, the local seed companies in Pakistan introduced BG-I into local varieties through 'introgression breeding'. Exotic varieties that were introduced directly were not performing well in Pakistan due to CLCV and high temperatures. The Agriculture Department was unable to effectively enforce or implement any refugeprogram or resistance management strategy. GMO technologies other than BG-1 were patented and technology providers were reluctant to come to Pakistan because the country lacked legal protection or plant breeder rights (PBR) until 2016. The multiple gene varieties developed by the local biotechnology institutes especially Centre of Excellence in Molecular Biology (CEMB) at Lahore could not be approved in Pakistan due to regulatory issues and the 18th constitutional amendment which placed agriculture in the provincial domain in 2012. These were a few of the chain events that resulted in the surge of PBW problem in Pakistan

Have there been any recent innovations for PBW management from research institutes in Pakistan?

Different groups are working on rearing PBW on an artificial diet developed by local ingredients for resistance monitoring studies. Some scientists are exploring the most effective recommendations of appropriate insecticides. Studies are underway for off season management of PBW at ginning factories and the impact of turning stored cotton-stalk stacks upside down two three time during season. Studies also showed that banning early sowing or fixing of an early crop sowing window combined with a mandatory installation of PB rope in early sown cotton crop, were found to be very effective. But all these strategies need a commitment and effective administrative follow up and coordination with district management.

Figure 3. Ginning mill waste with PBW larvae

Is there any concerted mission programme or a campaign to control or eradicate PBW in Pakistan?

As of now there are no mission mode programmes on a very big scale, but a program to introduce PB rope for PBW management has been in place for the last three years, albeit at a very limited scale for demonstration purposes. For off season management, the district governments introduced Section 144 and mandated ginning factories to dispose of mill waste, turn cotton stalk stacks upside down at fortnightly interval and enforce a restriction on early sowing within a designated window. Recently locally developed triple gene (Cry1Ac-Cry2Ab-EPSPS) varieties, CEMB Klean, Cotton 3 and CKC-3 were approved by the Provincial Seed Council. The National Seed Council allowed a fast-track procedure to approve some more promising varieties with the new technologies developed by local institutions in public and private domain.

Figure 4. Residual boll picking machine developed by CCRI Multan

Figure 5. A field after final picking. Left: stalks with left over bolls. Right: bare stalks after picking residual bolls with the boll picking machine

The Central Cotton Research Institute has re-engineered a boll picking machine, which picks up the left-over bolls (mostly infested) when operated after final picking. It collects more than 95% of the leftover bolls in a bin. The collected bolls are spread in sunlight for opening. Pheromone traps are placed nearby to catch any emerging adults from such bolls. Un-infested bolls do open and provide about 40-60 kg lint per acre. Though the lint is of inferior quality, it gives an additional income to farmer. The cotton stalks collected from the fields subsequent to the machine-pass, can be safely stacked and stored for fuel purpose without the fear of pest carryover from residual diapausing PBW larvae. A prototype machine was tested on farmers' fields and on campus last year and proved effective. Multiple machines are being prepared through a project to be used on a rental bases in cotton growing regions.

What in your opinion are the most important management strategies?

Looking into the sustainability and economics, I feel that an integrated approach would solve the problem. Importantly, off-season management through regulatory enforcement, is imperative to halt the population build up early in the season. Effective monitoring through moth-catches in traps and boll dissection is important to arrive at reliable economic threshold levels (ETLs) so as to take pesticide application decisions. Larvae in ginning waste can be used for multiplication of natural enemies in fields using Natural Enemies Field Reservoir (NEFR) Technology, which proved its effectivness in mealy bug management as well. Seed processing technologies can also play their role in PBW management. Sole reliance on synthetic pesticides triggers multi-dimensional problems that are difficult to manage and raise the cost of production. Chemicals be used as a part of strategy but should not become the only strategy. Pest scouting must be launched as a national campaign by mobilizing support from the civil society. PBW monitoring is a technical subject and the public sector can conduct capacity building programmes and data analysis for area specific recommendations. Well-designed strategies considering local agroecological conditions and social norms would be the best strategies for PBW management.

Biological Control and Pheromone Technologies will be Crucial for PBW Management

Interview with Dr Mohamed Negm

Dr Mohamed Negm is Professor of cotton technology, Cotton Research Insatiate, Giza-Egypt. Dr Negm is the Chair of the Research Network on Cotton for the Mediterranean & Middle East Regions-ICAC; Chairman of International Cotton Researchers Association-ICRA. Dr Negm filed a patent on: Developing a DNA-Based Technology for Identifying the Presence of Egyptian Cotton Fibre in Various Textile Products. He has special expertise in cotton and textile sectors, measurement and interpretation of fibre properties, marketing issues, processing efficiency, quality control and policy. Dr Negm received 'Encouragement Nation Award 2005', in advanced science technology, conferred by the Academy of Scientific Research and Technology.

How serious is the problem of pink bollworm in different cotton growing regions of Egypt?

The pink bollworm *Pectinophora gossypiella* (Saunders) (Lepidoptera: Gelichiidae) is considered one of the most important pests infesting the cotton crop in Egypt, because it is difficult to control with insecticides. Many eggs are laid on the sutures or under the bracteoles at the base of the boll, particularly on 14 day old bolls. Neonates penetrate flowers or bolls within 20–30 min.

What could the range of economic damage (%) be in different regions?

In Egypt, we have two regions: Delta Egypt (northern Egypt) and Upper Egypt (southern Egypt). This pest is very active in both regions. Larvae mostly remain inside the squares, flowers and bolls and cause severe damage, that may vary from year to year but generally cause 10-15 % of yield reduction in case of IPM, and with poor IPM it could reach 30%.

How serious was the pink bollworm in earlier times compared to what it is in recent years?

In earlier times, the Egyptian cotton leafworm, *Spodoptera littoralis* (Boisduval), was the most serious pest of the Egyptian cotton crop more important than the *Helicoverpa armigera* and *Earias* species. In the early 1980s, the government of Egypt prohibited child labour in cotton fields. Earlier children were employed for hand-picking of egg masses as a type of cultural control.

What do you think are the factors that prompted its recent resurgence as a serious pest?

Replacement of a primary pest with a secondary pest occurs when an insecticide treatment controls the primary pest and also destroys natural enemies of an injurious insect that was regulated below an economic injury level by the natural enemies, thus elevating the secondary pest to primary pest status.

Have there been any recent innovations for PBW management from research institutes in Egypt?

Eradication programmes using a combination of sex pheromone monitoring and mating disruption, irradiated sterile moth releases, and local insecticide application are the available options for the management of PBW infestation. However, the continued threat of reinvasion reinforces the need to improve pheromone-based monitoring. The egg parasitoid, *Trichogramma evanescens* West. has been used successfully for controlling the cotton bollworms, pink and spiny bollworms in cotton fields in Egypt.

Emphasis has been placed on biological control for the following reasons:

- Biological control can establish itself to provide season long control. The plant protection measures are eco-friendly in nature, and benefit farmers by reducing their investment on repeated insecticide sprays, with no adverse effect on the beneficial insects.
- The philosophy of protective biological control ensures the presence of a protective mechanism at the proper time before the bollworm larvae enter the fruiting parts of cotton plants. Once the larvae enter the bolls, they are protected from extraneous insecticide effects.

Is there any concerted mission programme or a campaign to control or eradicate PBW in Egypt?

In Egypt, the Academy of Scientific Research and Technology have announced an annual National campaign for cotton development that includes agricultural practices, irrigation, plant protection and gentle picking.

What in your opinion are the most important management strategies?

- The best option is the development of tailor-made systems for targeted management in affected areas with the selection of IPM components based on the PBW population density, crop production methods and economic feasibility.
- The potential long-term benefits of PBW population suppression on an area-wide basis appear to justify area-wide efforts in terms of reduced costs, more effective control, less environmental contamination and other peripheral problems associated with conventional control approaches.
- The unlikelihood of eradication indicates the need for long-term monitoring and programme maintenance following successful area-wide management.
- The success of area-wide PBW management is highly dependent on participation in the planning, site selection, implementation and assessment phases of the programme by all segments of the agricultural community.
- A highly effective extension and education communication programme is an essential component. Local uncoordinated efforts have not reduced the economic status of this pest in any area where it is an established pest.

Boll rot in a PBW affected boll. Though a larva feeds on a single loculi after entering the boll, it's entry hole makes the entire boll vulnerable to fungi especially under humid conditions.

Photograph: Mahesh Upender

PBW is Not a Major Issue in Greece

Interview with Dr Stefanos Andreadis

Dr Stefanos Andreadis is a researcher at the Institute of Plant Breeding and Genetic Resources of the Hellenic Agricultural Organization – DIMITRA, Thermi, Greece. His research field is on agricultural entomology. He is an active member of the Hellenic Entomological Society for more than 10 years and in the current period 2020-2021 he serves as a vice-chair. He authored two University textbooks and more than 40 peer reviewed research papers. Dr Andreadis is keenly interested in investigating the behavior and chemical ecology of tripartite relationships between insects, plants, and microorganisms with the ultimate goal to develop effective novel tools that attract and trap insects in terms of integrated pest management.

How serious is the problem of pink bollworm in Greece? What is the estimated range of economic damage (%)?

Nowadays, pink bollworm is not a serious threat for Greek farmers. Only occasionally and in specific areas there might be some issues, but in general the situation with PBW is "under control". The estimated range of economic damage is less than 5%

How serious was the pink bollworm in earlier times compared to what it is in recent years?

Indeed, the PBW in earlier times (i.e., 15 years ago) was a major issue. The estimated range of economic damage at that time was as high as 15-20%. Nowadays, as mentioned already, it is not considered as a major issue anymore. However, another lepidopteran species *Helicoverpa armigera* has taken over PWB causing big losses to the farmers, which sometimes can be as high as 50%. Almost every alternate year farmers are having to face great losses due to the attack of *Helicoverpa armigera*.

Have there been any recent innovations for PBW management from research institutes in Greece?

As far as I know, no there aren't any recent innovations for PBW management from research institutes in Greece. This is due to the fact that PWB is not a serious threat anymore.

Is there any concerted mission programme or a campaign to control or eradicate PBW in Greece?

Yes. there is an annual IPM mission programme that is utilised in the areas (Prefectures) where cotton is cultivated. This programme is a managed by state authorities and relies on the monitoring of all major pests of cotton and where appropriate the recommendation of using chemicals for control of PBW. This programme appears to work well in the case of PBW.

What in your opinion are the most important management strategies?

Monitoring PBW populations with pheromone-based traps for early detection and thereafter recommendation for any appropriate control measure (such as use of chemicals) in terms of IPM are the most important strategies

Short Season Compact Genotypes Planted at Higher Density Will Hold Fort Against PBW in India

Interview with Dr Y. G. Prasad

Dr Y. G. Prasad is currently working as Director, ICAR-CICR. He is an alumnus of Bapatla Agricultural College and earned his PhD in Entomology from the Indian Agricultural Research Institute. He worked extensively on development of microbial biopesticides, bio-ecology of invasive cotton mealybug, pest forecast research and development of decision support systems for pest management, climate resilience and agricultural extension.

How serious is the Problem of Pink bollworm in different Cotton growing States of India?

The first report of field-evolved resistance development by pink bollworm (PBW) to Bollgard (Cry1Ac) in India came in 2010 and subsequently to Bollgard II (Cry1Ac and Cry2Ab) in 2014. During the last 5 to 6 years early incidence of PBW was noticed on BG-II hybrids in the largest cotton growing tract in Central and South India. At present, PBW has become a key production constraint in the major cotton producing states like Maharashtra, Gujarat, Madhya Pradesh, Telangana, Andhra Pradesh and Karnataka together accounting for 85% of the cotton area in the country and the problem is expanding to newer areas as well.

How serious was the Pink bollworm in earlier times compared to what it is in recent years? What could the range of economic damage (%) per cent be in different States?

Pink bollworm infestation is showing an increasing trend late in the season during November to December and January months based on monitoring data of live larval recovery in random samples of green bolls collected from Bt-cotton fields across the country. During 2010 and 2011, pink bollworm incidence in green bolls was sporadic and was restricted mostly to non-Bt cotton in 11 out of 19 districts surveyed at that time. Surveys conducted every year after 2014 indicated higher incidence of pink bollworm larvae in green bolls collected from Bt cotton fields as well with larval recovery on BG-II averaging between 37 to 55%. Since 2017, surveys conducted late in the season (November onwards) indicated larval recovery of over 70% in sampled green bolls. The estimated economic damage beyond 120 days after sowing is between 20 to 25% given that mostly one or sometimes two locules are found infested in bolls. In the recent years, crop extended beyond December-January suffered 100% PBW infestation in developing bolls.

What do you think are the factors that prompted its recent resurgence as serious pest?

Poor adoption of refuge strategy in Bt cotton triggered the initial development and later widespread expansion of resistant populations of PBW in the largest cotton growing belt of central and south India since the outbreak in 2015 that was reported from Gujarat, accentuated the problem. The problem of PBW exacerbated due to several factors including adoption of a large number of long duration hybrids with varying growth patterns providing continuous flowering and fruiting periods for early appearance of the pest and development of overlapping generations late in the season. A cotton crop-free period of over 5 to 6 months after January until the next sowing in June is known to reduce infestation. In rainfed areas, farmers extend the crop duration to realize additional yield because it involves no extra cost. In certain years, this habit of extending the crop is reinforced as yield from later pickings compensates the loss incurred during first-second pickings. Poor retention of early formed bolls during the season due to weather vagaries in August-September and damage due to excess rainfall events in October is partly responsible for holding on to this practice. However, this exigency has become a perfect recipe for exacerbation of this late season pest.

Have there been any recent Innovations for PBW Management from research Institutes, All India Coordinated Cotton Improvement project and Universities in India?

Crop window-based pest management has been formulated by ICAR-CICR and advocated by the national agricultural research system. This comprises of several steps of crop regulation and management right from the time of sowing to harvest combined with monitoring of pest arrival and threshold-based treatment options at different crop growth stages. Pheromone technology for monitoring in the early to mid-crop growth stages and mass trapping and mating disruption options late in the season have been evaluated by several researchers. The latter option is likely to have better impact when practiced on area-wide basis. ICAR-CICR, Nagpur has developed and commercialised an innovative slow-release pheromone formulation with extended field performance. Eco-friendly treatment options such neem application for oviposition deterrence and population reduction through egg parasitisation by Trichogramma bactrae have been included in the management package for PBW. The practice of cultivating short duration and early maturing genotypes amenable for single picking sown under high density planting system has the potential of tackling the PBW menace which is most damaging late in the crop season. Any mismatch between susceptible crop stage and arrival of the pest is key for bringing down resistant populations over a period of time. Both public and private sector research is gearing up to put this approach to test. The All India Coordinated Cotton Improvement Project (AICCIP) has sponsored multilocation testing of early maturing varieties or hybrids paving the way for their testing and commercial release. ICAR-CICR undertakes regular resistance monitoring studies in field populations of bollworms to Bt Cry toxins (Cry1Ac and Cry2Ab). Weekly advisories and pest alerts (text/ voice messages) are issued in local languages to farmers and State Departments.

Is there any concerted mission programme or a Campaign to Control or Eradicate PBW in India?

Area-wide management of the pest warrants community action in a campaign mode. Awareness creation on management actions at different crop growth stages during the season starting from sowing to harvest followed by crop termination, crop residue management and trapping of adult moths at ginneries are crucial factors in the fight against PBW. ICAR-CICR was the first to advocate a stepwise pest management package that was first implemented in Gujarat state and later in Maharashtra state in campaign mode. The insecticide (Bt) resistant management (IRM) project is being implemented in all the 11 cotton growing states in 21 key districts to create awareness and disseminate IPM measures for PBW management since 2018. Eradication of the pest is challenging due to widespread nature and prevalence of mostly resistant populations. The success achieved through sterile male release technique in USA may not be relevant now to Indian conditions for this reason.

What in your Opinion are the most important management Strategies?

A host of interventions are needed to keep in PBW in check. Adoption of short duration compact genotypes planted at higher density and amenable for single picking is a key strategy for PBW management in India. Monitoring, mass trapping and mating disruption using pheromone technology are likely to help reduce population levels and economic loss. Use of neem and biological control agents at flowering along with need based use of insecticides at boll development stage may help minimise cost of cultivation to a certain extent and maximise economic returns.

Photograph: Keshav kranthi

Pink Bollworm Resistance to *Bt* Cotton and Management Considerations

Dr G. M. V. Prasada Rao, Dr Sandhya Kranthi[1] and Dr Keshav Kranthi[2]

Dr G. M. V. Prasada Rao, PhD, Principal Scientist (Entomology), ANGRAU, LAM, Guntur, Andhra Pradesh, India, has 20 years of research and extension experience in cotton. He published 60 peer-reviewed research papers and four books/manuals. Dr Prasada Rao bagged 16 recognition awards including the prestigious National Award "Dr. Bap Reddy Memorial Award" for contributions in the field of IPM. He handled 200 lakhs worth externally funded research and extension projects. Dr Prasada Rao guided ten postgraduate and Ph.D. students in cotton Entomology. As a Course Director, he conducted an ICAR sponsored Winter School on IRM. Dr Prasada Rao is a member of ten professional scientific societies and the Fellow of Plant Protection Association of India.

Pink Bollworm Incidence Was Low in India Before the *Bt* Era

Interview with Dr GMV Prasada Rao

How serious is the problem of pink bollworm in AP? What could be the range of economic damage (%)?

Andhra Pradesh is one of the important cotton-growing southern states in India. The Pink bollworm, *Pectinophora gossypiella* attacks cotton late in the season. Of late, the PBW has become a major production constraint on cotton. Yield loss ranged from 20-50%. Around 50% damage to seed cotton was observed in some intensive cotton-growing blocks or mandals of the Guntur, Krishna, and Prakasam district of AP during 2020-21. Further, the damage was observed in October-November months onwards itself (2nd picking).

How serious was the pink bollworm in earlier times compared to what it is in recent years?

Pink Bollworm incidence was low before Bt era. Before *Bt*, the incidence was low in Andhra Pradesh. After *Bt* also till 2007-08, incidence was low and economic damage was not much since it was causing sporadic damage to final harvests. From there to 2014-15 incidence was almost nil in AP. However, since 2015-16 season again it started appearing and caused severe damage during 2017-18 and 2020-21.

What do you think are the factors that prompted its recent resurgence as a serious pest?

- Widespread cultivation of long staple *G. hirsutum Bt* hybrids (>99% of the cotton area is under *Bt* hybrids).
- Continuous sowing and availability of cotton in different regions of AP (Round the year)
- Non-practice of stipulated IRM/IPM strategies
- Development of resistance in PBW against *Bt* hybrids

What could have prompted the rapid development of PBW resistance to Bt-cotton?

- High selection pressure due to extensive cultivation of only one or two *Bt* events.
- Monophagous nature of the pest
- Non-practice of the refuge strategy

Have there been any recent innovations for PBW management from research institutes?

- Efforts are in progress. Nonetheless, results of on-farm trials on Mass-trapping indicated positive in management of the pest. Likewise, mating disruption technology also promising.

What in your opinion are the most important management strategies?

- Practice of Dead or Closed period for 120-150 days (practically no cotton cultivation between seasons)
- Aggressive promotion of Pheromone technology: Mass trapping / Mating disruption
- Strict implementation of IRM/IPM on a community basis involving all the stakeholders.
- Cultivation of different short duration *Bt* cotton varieties/hybrids.

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Introduction

Genetically modified cotton (*Bt* cotton) was cultivated in 12.3 million hectares (ha) in India during 2020, which is 95% of the total cotton area of 12.9 million ha grown in the country. An estimated 9.8 million cotton farmers adopted *Bt* cotton technology. Studies show that *Bt* cotton is still providing good protection against *Helicoverpa armigera* and *Earias* spp. in the country. The studies also align with the fact that as of now there are no field level complaints of economic damage or development of practical resistance in *H. armigera* and *Earias* spp to *Bt* cotton in India. However, increase in resistance allele frequency to Cry1Ac was documented in field-collected population of *H. armigera* from Telangana and Andhra Pradesh in India. The combined allele frequency during 2013 and 2014 was 65 times and 29 times higher than the initial frequency recorded during 2004 (Kukanur *et al*., 2018), respectively. On the other hand, severe field incidence of pink bollworm (PBW) was noticed in some central (Gujarat and Maharastra) and Southern (Telangana and Andhra Pradesh) cotton-growing regions (Kranthi 2015 and Mohan 2017). Dhurua and Gujar 2011 confirmed field-evolved resistance to Cry1Ac in PBW populations collected from Gujarat. Further, Chinnababu Naik *et al*., 2018 conducted extensive resistance monitoring studies in PBW populations from 2010-2017 in 38 districts of 10 major cotton growing states of India and concluded that the PBW populations developed resistance to Cry1Ac and Cry2Ab in major intensive cotton growing districts of Central and South India. The *Bt*-resistant pink bollworm larvae have been causing considerable economic damage that led to panic in the Indian and Pakistani cotton sectors. The concerns were serious because not only did yields decline but the fibre quality deteriorated significantly enough to warrant a reduction in the market price of the 'poor quality PBW damaged cotton'. Strangely, this problem is unique to India and Pakistan and not in any other *Bt* cotton growing country in the world. Interestingly, all major cotton growing countries have been able to control PBW with *Bt*-cotton. Tabashnik and Carriere, 2019 reviewed the global resistance monitoring data and indicated that resistance management strategies adopted by three major cotton growing countries in the world; the USA, China and India had significant impact on the interaction of *Pectinopora gossypiella* with *Bt* cotton. Integrated Pest Management holds a crucial role in the management of this important pest on cotton in India (Kranthi, 2015 and Tabashnik, 2019). This paper discusses the status of insect resistance to *Bt* cotton in India and possible reasons for the development PBW resistance in India — why the situation is unique to India — and discussed different Integrated Pest Management recommendations for sustainable cotton production India.

Status of insect resistance to *Bt* cotton in India

Helicoverpa armigera

Kranthi (2012) reported that resistant ratios up to 31-fold were recorded in the populations tested from 2008/09 to 2010/11; 51-fold in one location from 2011/12 and 128-fold in two locations in 2016 (Kranthi, unpublished data). He stated that, *Bt* cotton continued to be effective against *H. armigera* even in those regions where highest RRs of 128-fold were observed. Survival of *H. armigera* populations on *Bt* cotton and field failures were not observed from any cotton growing region of India. Kukanur *et al*., 2018, reported an increase in resistance allele frequency to Cry1Ac in field collected populations of *H. armigera* from southern states of India; Telangana and Andhra Pradesh. The combined allele frequency during 2013 and 2014 was 65 times and 29 times higher than the initial frequency recorded during 2004, respectively. Singh et al (2021) reported Cry1Ac resistance allele frequencies of 0.050 (95% CI 0.022–0.076) and 0.056 (95% CI 0.035–0.075) in *Helicoverpa armigera* populations collected from pigeon pea grown alongside *Bt* cotton in 2016 and 2017 in the Telangana state of India. They stated that the resistance allele frequencies to Cry1Ac in the cotton bollworm remained unchanged compared to their earlier studies conducted in 2013 and 2014. Thus, Indian populations of *H. armigera* appear to be still susceptible to *Bt* cotton (Cry1Ac) at the field level.

Pectinophora gossypiella

Chinnababu Naik *et al*., 2018 reported negligible larval incidence of PBW on *Bt* cotton in North India. But, in Central and South India larval recovery in *Bt* cotton ranged from 29 to 72% during 2014-2017. Likewise, the mean Resistance Ratio (RR) for Cry1Ac was 47 (18-127) during 2013 and increased to 1387 (704-2060) during 2017. A similar increasing trend was observed for Cry 2Ab with a mean RR increase from 5.4 (1-31) in 2013 to 4196 (1306-9366) in 2017.

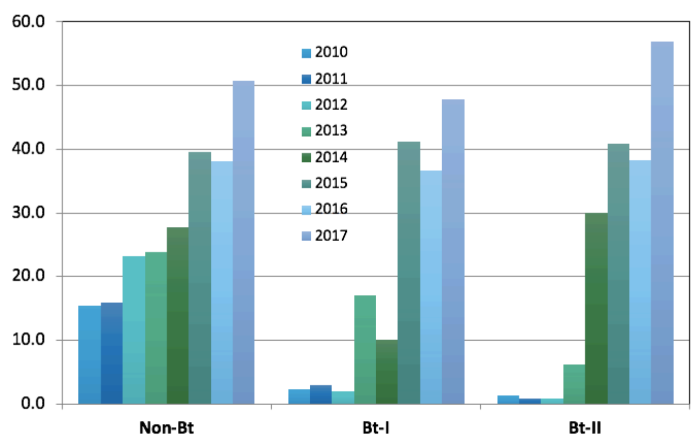

Figure 1. Increased levels of PBW infestation from 2010 to 2017 due to resistance development

Tabashnik and Carriere, 2019, reviewed the global resistance monitoring data and indicated that resistance management strategies adopted by three major cotton growing countries in the world; the USA, China and India had significant impact on the interaction of *Pectinopora gossypiella* with *Bt* cotton. They primarily pointed that abundance of refuge varied among these three countries that might have played a key role in the striking differences in the incidence of the same pest species on the same crop and on the same toxins, without discounting the role of other differing factors like nature of hybrids and varieties, climate and production practice adopted in the three major cotton growing countries in the world. Further, PBW populations in all the three countries contain cadherin mutations responsible for Cry1Ac resistance (Morin *et al.*, 2003, Fabrick *et al*., 2014 and Wang *et al*., 2019) and ABC transporter mutations in population of the USA and India conferring resistance to Cry2Ab (Mathew *et al*., 2018).

Possible causes for the recent PBW resurgence

The emergence of PBW as a monstrous pest of cotton in recent years in India is unprecedented. PBW was a serious pest for a long time in India mainly because of the long season which enabled the pest to thrive for several overlapping cycles, multiply and reinfest. The pest was relegated to a minor status soon after 1980 with the advent of short season varieties coupled the introduction of synthetic pyrethroids. After a hiatus of almost 30 years, PBW emerged as a major pest again after 2010, and this time more severely on *Bt* cotton, which was supposed to keep it under check. Its reappearance as a major pest in recent times appears to have been a result of six major factors (Kranthi, 2015) that are listed below:

Development of PBW resistance to *Bt*-toxins

Pink bollworm populations developed resistance to Cry1Ac and Cry2Ab and are thus able to survive and multiply on *Bt*-cotton to cause extensive crop damage.

The use of *Bt*-hemizygous hybrids

Studies (Kranthi, unpublished) showed that PBW survival was higher in *Bt*-hemizygous hybrids compared to *Bt*-homozygous varieties. This could have been due to low toxin expression and the presence of Bt-toxin segregation in developing bolls. The *Bt*-hemizygous hybrids had a low dose of *Bt*-toxins in developing fruiting parts thereby accelerating resistance. Younger developing stages such as square buds, flowers and developing younger bolls were found to have low *Bt* toxin levels that may have helped the heterozygous-resistant PBW to survive better. The seeds present in bolls of the *Bt*-hemizygous F-1 *Bt* cotton Bollgard-II (2-gene) hybrids segregate in 9:3:3:1 ratio for the cry1Ac and cry2Ab genes. The segregation ratios mean that about 6% of seeds do not contain any *Bt* toxin, 56% of seeds contain a mix of Cry1Ac and Cry2Ab, 19% of seeds contain Cry1Ac and the rest of 19% contain Cry2Ab. Thus, the seeds in a single boll at a time contain the two toxins in different ratios and different toxin titres (Kranthi, unpublished data) that helps PBW larvae to survive and develop resistance more rapidly because of the possibility of heterozygous-*Bt*-resistant larvae being able to survive on non-*Bt* seeds and the low-dose toxin seeds.

The return of long season cotton

The introduction of long season *Bt*-cotton hybrids provided a long season survival opportunity. The long season hybrid cotton varieties served food for PBW survival directly for a long period over the season and the multiplicity of hundreds of *Bt*-cotton hybrids which have overlapping flowering and fruiting cycles especially when sown in a long-staggered sowing window helped the pest to survive additional cycles that resulted in higher pestilence. A large number of hybrids with varying flowering and fruiting regimens provide continuous food for PBW in an overlapping manner over a long period to enable a greater number of generation cycles in the longer season, and thus a larger residual population flow into the subsequent seasons. Early sown (April-May in central India) crop in some regions starts flowering and reaches a peak flowering stage that coincides with the first minor seasonal PBW peak pink bollworm that occurs in July, thereby providing food for an otherwise suicidal population. Almost all the *Bt*-cotton cultivating countries in the world cultivate Bt cotton varieties except India which grows *Bt* cotton hybrids, that are grown for a longer duration of 180 to 240 days, thereby serving as continuous hosts for PBW.

Lack of adequate refuge hosts

The recommendation of 20% non-*Bt* cotton to be grown as five border rows was not easily accepted by Indian farmers because of poor quality non-*Bt* seeds supplied by many seed companies and the fear of suffering losses due to bollworm infestation in the 20% refuge non-*Bt* crop. In hindsight, it appears that the recommendation was not practical. PBW is functionally monophagous on cotton with few alternative hosts in the cotton cropping systems. Therefore, resistance development seems to have been inevitable due to a strong selection pressure in the absence of non-*Bt*-cotton or other non-*Bt* host crops that may have otherwise slowed down resistance development as refuges.

The synthetic pyrethroid factor

It is widely believed that the introduction of synthetic pyrethroids in 1980 resulted in a significant decline in the PBW pest status in India. PBW showed signs of resistance to synthetic pyrethroids by mid-nineties, but not strong enough to warrant control failures. Bollworm resistance to pyrethroids and introduction of *Bt*-cotton led to a decline in insecticide usage during 2002 to 2007, especially to a significant reduction in pyrethroid usage across India which may have allowed the pest to survive better.

Ignoring IPM

Bt cotton is meant to control all three bollworms. Farmers were told that the technology would effectively control all three bollworm species including pink bollworms. Thus, there was a general notion that bollworms would be controlled by *Bt* cotton and farmers were expected to take care of sap-sucking insect pests. Consequently, integrated pest management (IPM) methods were mostly oriented toward sucking pests and not toward bollworms. In effect, IPM for bollworms was completely ignored. It was only after the emergence of PBW resistance to *Bt* cotton that seed companies and the technology developers of *Bt* cotton started reiterating the importance of IPM in bollworm management.

Why pbw resurged in India & Pakistan but not in China & USA

Pink bollworm is a global pest. While PBW emerged as a major pest recently in India and Pakistan, it continues to maintain a low profile in other countries where it was once a very serious pest. India opted for *Bt*-cotton hybrids, whereas Pakistan like rest of the world chose to grow *Bt*-cotton open-pollinated varieties (OPV). India grows Bollgard-II® *Bt*-cotton which has two *Bt*-genes *cry1Ac* and *cry2Ab*, whereas Pakistan continues to depend on Bollgard® *Bt*-cotton which expresses a single *Bt* gene *cry1Ac*. But despite the differences in hybrid v/s OPV and the single gene v/s two genes, PBW emerged as a major pest in both countries.

Two main factors may have influenced higher levels of PBW pestilence in India and Pakistan. PBW survives better in both countries because it developed resistance to *Bt*-cotton. The second factor pertains to the long season cotton in both countries. While *Bt*-hybrids grown at a low density in India warrant a longer season for a harvest of decent yield, *Bt*-varieties grown a higher density as in Pakistan do not need to be grown over a longer season. However, the sowing window in Sindh province of Pakistan extends over a long period of almost four months from mid-March to mid-June. Thus, Pakistan has a shorter gap of only three to four months of a closed-season or a cotton-crop-free period; a situation that is similar to India.

The cotton crops in USA, China, Brazil, Australia, Turkey etc., are grown for five to six months, thus allowing a 'closed-season' of six months.

USA

The United States initiated a successful science driven campaign to eradicate pink bollworm. The cotton growing season continues to be restricted to 5-6 months. Apart from implementing a suite of cultural methods, scientists helped farmers ensure a strong compliance of non-*Bt* refuge cultivation. Scientists also coordinated mass releases of sterile PBW moths that helped to minimise populations and deployed pheromone technology for monitoring and mating confusion. USDA worked closely with the Mexican government to implement the programme jointly. Mexican Government agencies (SAGARPA and SENASICA) and United States (USDA) together with scientists chalked out plans meticulously and executed them to near perfection over fifty years to eradicate the pink bollworm. The key components of the eradication programme were pest surveys using delta traps, sterile insect technique, pheromone-based mating confusion, short season and transgenic Bt cotton. Mating confusion was through PBW Rope technology used at 500 ropes per hectare as one or two applications or 4-5 pheromone sprays of NoMate or CheckMate per season. Two sterile moth rearing facilities were established in Harlingen, Texas and Phoenix, Arizona in 1968 and 1969 and a new facility in a building of 6131 square metres was developed in Phoenix in 1995. Sterile insect releases started in 1968 in California and have continued ever since. It is worth mentioning that by 2005, the Phoenix rearing facility was producing 20 million to 28 million moths per day. The sterile moths were labelled with Calco red oil food dye or strontium and tracked diligently. Thus, cotton remains PBW-free in the USA because of a combination of IPM and IRM technologies. Bt-cotton technology also played a major role in PBW eradication.

Bt-cotton varieties in the USA express a range of *Bt*-toxins which makes it relatively difficult for bollworms to develop resistance to all the *Bt*-toxins. The availability of different *Bt* cotton varieties for technology durability and improved insect resistance; Bollgard® 1 containing Cry 1Ac, Bollgard® II with Cry1Ac + Cry2Ab, Widestrike™ with Cry1Ac + Cry1F, Widestrike® 3 contains Cry1Ac, Cry1F and VIP3a, Bollgard® 3 containing VIP 3a along with Cry1Ac and Cry2Ab, Twinlink® Cry1Ab and Cry2Ae, Twinlink® plus containing Cry1Ab, Cry2Ae along with VIP3Aa19.

China

In China, farmers continue to depend on cry1Ac based *Bt*-cotton which in some varieties is pyramided with a protease inhibitor gene. Bollgard II cotton is not grown in China. Farmers cultivated F_2 hybrid seeds from crosses between *Bt* and non-*Bt* cotton, producing 25% non-*Bt* plants which acted as refuge in *Bt* cotton. Seed mixtures generated with F_2 hybrids in China were found to have been effective in delaying PBW resistance development against *Bt*-cotton. In 2011-2015, F_2 hybrid fields accounted for a mean of 67% of the total cotton cultivated and PBW population was reduced by 96% & insecticide sprays were reduced by 69% compared to 1995-1999 (non-*Bt* era) (Wan *et al*., 2017). Further, cultivation of short season cotton also might have a role in delaying the development of resistance to the pest (Dai and Dong 2014).

PBW management strategies

A range of management strategies, especially for Indian conditions, have been proposed by Kranthi (2015). A short season coupled with a closed season of 5-6 months plays a crucial role in the management of cotton bollworms especially pink bollworm. Other technologies such as pheromones for monitoring, mating confusion and mass trapping, crop residue management, judicious nitrogen usage and diligent insecticide management contribute to minimise PBW populations and damage.

Fertiliser management

A long season crop is most vulnerable to PBW infestation. Excessive nitrogenous fertilizers create conditions for a long season crop through a combination of two effects. The first effect is direct wherein excessive nitrogen leads to excessive vegetation and delayed maturity. The second effect is indirect wherein excessive nitrogenous fertilisers cause higher susceptibility to sap sucking insects, which warrant the use of organophosphate and neonicotinoid insecticides, most of which cause delayed maturity of the crop. It is important to practice balanced application of NPK; avoid excess use of nitrogenous fertilisers and encourage the use of organic fertilizers.

Pesticide management

A few types of organophosphate and neonicotinoid insecticides are known to cause physiological changes in plants to delay crop maturity. Late maturity extends the season thereby enhancing vulnerability to PBW infestation and damage. Organophosphate and neonicotinoid insecticides are used for sucking pest management early in the season. Selection of sucking resistant/pest tolerant hybrids, helps to avoid spraying of insecticides such as monocrotophos, acephate, imidacloprid, thiomethoxam etc., during the early crop growth period; these chemicals delay crop maturity. Avoiding these chemicals helps in synchronous early maturity of the bolls resulting in less incidence of PBW. Though synthetic pyrethroids are effective in controlling PBW, their indiscriminate use can lead to resurgence of whiteflies and American Bollworm. Therefore, avoid spraying of synthetic pyrethroids until the economic threshold levels (ETLs) for PBW are reached late in the season. ETLs of 8 moths per pheromone trap per three consecutive nights are used for insecticide interventions. Though conventional insecticides such as thiodicarb, profenophos, quinalphos or chlorpyriphos 20% EC @ 500 ml/ac are recommended, recent studies show that chlorantraniliprole, emamectin benzoate, flubendiamide, spinosad and indoxacarb are also effective and have a better environmental profile compared to the conventional insecticides.

Cultural practices

Several cultural practices have been reported to be highly effective in PBW management. Practices such as removal of ratoon plants or rogue plants; flooding of fields after harvest to kill residual diapausing larvae and pupae in soil; removal and destruction of rosette flowers, dropped squares and pre matured bolls; crop rotation and deep ploughing to expose PBW diapausing larvae and pupae to birds and excessive heat were found to contribute to enhancing pest control efficacy. Maintaining a non-*Bt* refuge crop helps in delaying the development of PBW resistance to *Bt* cotton.

Crop residue management

Cotton crop residues and ginning mill waste harbour pink bollworm larvae and pupae (Novo and Gabriel, 1994). PBW larvae enter into diapause and can be found in seeds and other crop residues. Non diapausing larvae become pupae and the adults that emerge propagate their generations on crops such as okra, hemp, roselle or weeds belonging to Malvaceae, Fabaceae, Convolvulaceae and Euphorbiaceae. Crop residue management and ginning mill waste destruction play an important role in minimising carryover populations of the pink bollworm. The majority of diapausing pupae are expected to be present in late season bolls, especially the abandoned last lot of immature bolls. Strategies such as defoliation, removal of late season green bolls and desiccation of the crop at the end of season were found to be effective in reducing over wintering larvae in the US (Adams, 1995). Encouraging the practice of diligent sanitation in ginneries and fields helps in reducing the residual pest carryover. Allowing cattle to graze in fields after final harvest was found to be beneficial because these animals were found to feed on immature green bolls that were the main sources of pest carryover.

Figure 2. Immature unharvested residual bolls in stacks of cotton stalks, crop debris on soil and diapausing pupae in seed cotton at gins are sources of PBW carryover. (Photos: Kranthi)

Pest-crop synchrony management

There are two theories —namely early planting and late planting — that have been proposed for PBW management to create asynchrony between pest occurrence its food. Early planting could ensure that bolls mature before PBW occurrence and escape, because peak populations of PBW are known to occur only late in the season. Though, PBW is a late season pest it also occurs as a small peak early in the season that acts as a precursor of the ensuing populations. Late planting has been suggested as a strategy so that the early emerging PBW populations do not have access to any food supply and starve to death (Frisbie et al., 1989). Thus, PBW infestations could be greatly reduced in the absence of the early precursor populations. In both cases, whether early of late planting, the strategy of deploying short-season varieties that are terminated in time and crop residues are desiccated has been acknowledged to be effective in minimizing pestilence and pest carryover. *Bt*-cotton was able to resist PBW damage until the emergence of insect resistance. A few varieties have been reported to have a higher tolerance to pink bollworms in China (Wu, 1993; Wang et al., 1993).

Biological control

There are two parasitoids that have so far been effectively used in PBW management across the world. The egg parasitoids *Trichogramma bactrae* or *Trichogramma brasiliense* have been recommended @ 60000 eggs per acre at peak flowering period to enable the egg parasitisation. *Bracon kirkpatricki* is a larval parasitoid that can play an important role in PBW management, provided the naturally occurring populations are allowed to establish and proliferate and parasite populations are also augmented if and when necessary.

Figure 3. *Apanteles* parasites recovered from field collected PBW larvae

Figure 4. *Bracon* parasites recovered from field collected PBW larvae

Figure 5. Deformed per-pupa and pupa

Closed Season

A closed season is legally enforced to prevent pest carryover to the subsequent season. For a closed season to be enforced, cotton plants must be destroyed to create a dead period in order to prevent build-up of pests. In countries like Zimbabwae, a closed season is governed by the Plant Pest and Disease Act, which stipulates that any farmer who fails to comply will face a fine or imprisonment or both (Mubvekeri and Nobanda, 2012). In the USA also, by the end of the 1980s, most of the cotton growers abandoned the cultivation of cotton in Arizona and California, the remaining cotton growers formed the Cotton Pest Abatement district and adopted a short season strategy and successfully managed pink bollworm on cotton (Thomas Miller, 2001). In India too, as early as 1911, cultural control in the form of removal of cotton sticks by 1 August every year was made compulsory by law to minimize incidence of pink bollworm on cotton in Madras State. Chinnababu Naik *et al*., 2018, attributed the 'Closed Season' as one of the major factors for maintaining susceptibility of PBW to *Bt* toxins in North Indian conditions. The cotton season in North India is restricted to 5-6 months to facilitate following wheat cultivation which creates a closed season, thus reduction in selection pressure because of a smaller number of PBW generations are exposed to the *Bt* toxins. However, cotton is now being cultivated over a longer season in parts of Pakistan, which might enable a higher survival rate in the bordering regions of India to elevate pestilence in north India.

Sampling and pest monitoring

Pink bollworm infestation must be closely monitored. Pheromone traps provide a reliable indication of the initial occurrence and continued infestation all through the season. Light traps also provide useful indications of pestilence. However, pheromone traps and light traps only capture moths but may or may not necessarily relate directly to the damage to squares, flowers and bolls. It is important to sample rosette flowers and green bolls regularly to assess the extent of damage and PBW population levels in the field so as to determine the best time to intervene.

Figure 6: Field monitoring and sampling

Pheromone technologies

Pheromones have been effectively used in PBW management by deploying them for monitoring, mass trapping and mating confusion. Lykouressis *et al*., 2005, evaluated the mating disruption of pink bollworm by monitoring its population with pheromone baited traps as well as sampling flowers and bolls to record damage levels in cotton fields during 1988 and 1989 in central Greece. The treated fields were compared with control fields in which 2-3 insecticide sprays were applied. In both years, the number of male moths caught in pheromone traps was greatly reduced in treated compared to control fields. Mating disruption reached 99.1%, 96.8% and 93.2% in different treated fields. In the treated fields, moth catches were reduced more in rows perpendicular rather than parallel to the prevailing wind. Staten *et al*. (1987) indicated that mating disruption greatly contributes to reducing the possibility of late outbreaks of secondary pests. Finally, it was concluded that mating disruption played a key role in reduction of pink bollworm catches in traps and lowering the damage. This effectiveness was significantly higher when planting lines were perpendicular to the direction of the prevailing winds. Damage levels were not proportionally reduced compared to the reduction of moth catches. Therefore, when mating disruption is adopted it must be accompanied by monitoring for damage levels. This clearly indicates the importance of this method in PBW management as part of Integrated Pests management (Cork and Hall, 1998). Jahnavi *et al*., 2019, reported that IPM module focusing on mass trapping of PBW resulted in less (10%) open boll locule damage due to PBW in IPM module over 24% in farmers' practice. Further, IPM module yielded 20% more seed cotton.

Conclusion

Pink bollworm is a pest that is difficult to control because it is an internal feeder and is not very vulnerable to biological control or chemical pesticides. However, it is clear from a global assessment that PBW became a major problem in countries such as India and Pakistan — countries where cotton is grown as a long season crop — whereas countries such as USA, China and Australia have either been able to eradicate it or prevent its emergence as a key problem by using a slew of IPM tactics such as short season, closed season, mating disruption and crop residue management. PBW is now a major problem in India and Pakistan. India is in a strange predicament in which 95% of its area is under hybrid cotton and the majority of cotton hybrids that are grown at low density cannot give high yields if terminated within 5-6 months; therefore neither is a short season nor a closed season possible. Pakistan is in a strange predicament because staggered sowing is followed in Sindh based on the availability of irrigation water. Staggered sowing leads to a long season that makes the crop vulnerable to PBW infestation and late season makes the crop more vulnerable to whitefly and the dreaded leaf curl virus. India and Pakistan must explore options to restrict the total area-wide cotton season within a state/province to less than six months and also to deploy strategies to retain early formed squares which enables higher yields from a short season. Other options related to fertiliser management, insecticide management, pheromone management, crop-pest asynchrony management, crop residue management, and cultural practices could lay a foundation for sustainable long term PBW management.

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The ICAC Recorder

An Update on the Biology and Natural Enemies of the Pink Bollworm

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Introduction

The Pink bollworm *Pectinophora gossypiella* (Saund.) (PBW) has emerged as a major problem for cotton production in India and China in recent years. Reports indicate that PBW resistance to Bt-cotton was one of the major reasons for the pestilence. Cotton cultivation in India faces an unusual challenge where the pest has evolved resistance to both Cry1Ac and Cry2Ab toxins resulting in field control failures of BGII cotton, while PBW has been reported to have developed resistance to Cry1Ac. Interestingly, PBW continues to remain susceptible to Bt-cotton in China, USA, Mexico and other countries where Bt cotton is grown. PBW has been the focus of attention across cotton growing countries across the world. Almost all countries have grappled with the pest some time or the other in the past hundred years. Many countries have devised long term management strategies and mandated a compulsory closed season to prevent carry over of the pest. USA and Mexico went a step further with pink bollworm eradication (Tabashnik, and Carrière, 2019, Tabashnik 2021) programmes that have been implemented since 1968. This article is an update on the biology and natural enemies of the pink bollworm. It also discusses the PBW management strategies adopted in non-Bt countries where it is currently not considered as a serious pest.

Biology

The most vulnerable stages of this insect pest to natural mortality are eggs, early instar larvae, and pupae in soil. A window of one week in the life cycle of the pink bollworm is available for parasitisation. Overlapping generations of the pest facilitate the availability of the vulnerable stages of the insect as food for natural enemies during various stages of the crop. Eggs are laid in green tissues near the reproductive parts of the plant, particularly on the bracts. Recent scientific papers reconfirm that

Duration of life cycle (days)	Egg period Days	Larval period (days)	Host	References
29-55.69			On artificial diet at 4 temperatures	El-Sayed, 2005.
		Dec-15	Cotton	Ellsworth et.al., 2006
		Oct-14	Cotton bolls	Vennila et al 2007.
		18.97	Wheat germ-based diet	Wu, et al 2008.
		18.7-27.56	On artificial diets with different seed powder sources	Muralimohan et al., 2009
35-40	03-May	Sep-13	cotton	Shah et al 2013
		18-19	Cotton varieties seed powder	Malthankar and Gujar,2014.
46-70	04-Jun	15-23	Artificial diet	Zinzuvadiya et.al., 2015
30-31			On Giza cotton	El-Lebody,et.al., 2015
	4.2-5.4	24-26	Artificial det	Dhara Jothi, et al 2016.
		24.7-25.9	Artificial diet	Sabry and Abdou 2016.
		7.76-30.89	Artificial diet	Massoud et.al., 2017
34-67			Artificial diet	Zinzuvadiya et.al., 2017
46.82	2.5-4.5	22.5-28.5	Bt cotton	Shrinivas et. al., 2019
40.58		20.5-24.5	Okra	
		15-18	Okra	Umer et al 2019
52.70			Okra	Mushtaq et.al., 2021
54.8			Cotton	

egg stage of the pink bollworm lasts for 2.5-6 days; larval period for 7.7 to 31 days and the complete life cycle could extend from 29 to 70 days. Larval duration and the total lifecycle of the pest is influenced both, by climatic factors and the host plant- cotton or okra. Climate change, particularly a rise in temperature, is expected to impact the life cycle and seasonal occurrence and abundance of the pest.

Natural enemies

The pink bollworm has a high host specificity and is the primary pest of cotton with very few alternate host plants. Its regulation by natural enemies is low, unlike the American bollworm, Tobacco budworm or the fall armyworm (Gutierrez et.al., 2006). The most vulnerable life stages to parasitization are the egg stage and neonates. Trichogrammatids parasitize the eggs while Braconids and Chalcids parasitize the larvae. Augmentation of egg parasitoids and conservation of larval parasitoids are popular methods of non- insecticidal pink bollworm management strategies. Predation of the egg, early larval and pupal stages has been commonly reported. Most predators lack morphological modification of mouthparts necessary to penetrate bolls to feed on pink bollworm larvae that reside inside bolls and feed. Pupation in the soil debris makes pink bollworm vulnerable to predatory ground beetles, Anthocorids, entomopathogenic nematodes and mirids. Natural enemies of the pest, mentioned in recent reports, are enlisted in Table 2. About 92 natural enemies of the pink bollworm from across the globe are listed in a data sheet available at https://www.cabi.org. Earlier reports (Branch, 1969; Jackson, 1978) list native natural enemies and describe the introduction of at least 14 endophagous insects into the US for pink bollworm management. Other references such as Cheema et.al., 1980, Greathead, 1989, Green and Lyon (1989) also enlist the natural enemies of this pest.

**Erigone atra, E. prominens, Erigonidium graminicolum, Gnathonarium gibberum, Oedothorax insecticeps, Enoplognatha dorsinotata, E. japonica, Theridion octomaculatum, Neoscona doenitzi, N. nautical, N. theisi, Singa hamata, S. pygmaea, Misumenopos tricuspidata, Synaema globosum, Thomisus labefactus, Xysticus croceus, X. lateralis atrimaculatus, Oxyopes sertatus, Pardosa T-insignita, Pirata japonicas, Chiracanthium japonicola, Clubiona japonicola, Marpissa magister, Plexippus setipes, Dyschiriognatha quadrimaculata, Tetragnatha cliens, T. extensa, T. japonica, T. nitens, T. praedonia, T. shikokiana, T. squamata*

NATURAL ENEMIES	EGG	LARVA	PUPA	ADULT	Reference
PARASITOIDS					
Trichogramma evanescence	■				Gergis 2004. Amin and Gergis, 2006, Saad et.al., 2012, Darwish et.al., 2017, El-Bassouiny,2021,
T. chilonis	■				Ahmad et.al., 2005, Sarwar 2017,
T. confusum,	■				
T.bactraea	■				El- Hafeez and Nada, 2000, Amin and Gergis, 2006, Sarwar 2017
Bracon gelechiae		■			Pathan et.al., 2019
Apanteles angaleti		■			
Dibrachys cavus		■			El-Sayyad, 2005
Brachymeria		■			
Bracon sp.		■			Sarwar, 2017, Naik et.al., 2018, Pathan et.al., 2019.
Chelonus curvimaculatus		■			Sarwar, 2017
Microchelonus blackburni.		■			Sarwar, 2017
Bracon lefroyi		■			Naik et.al., 2018
Chelonus pectinophorae,		■			Luo et.al., 2014
Bracon nigrorufum,		■			
B. isomera		■			
Goniozus legneri		■			El-Husseini et.al., 2018
G. pakmanus		■			
Steinernema carpocapsae		■			https://www.cabi.org/isc/datasheet/51706
PREDATORS					
Geocoris punctipes		■			Henneberry, 2007
Geocoris ochropterus,	■	■			Luo et.al., 2014
G. pallidipennis,	■	■			
Orius tristicolor		■			Henneberry, 2007, Luo et.al., 2014
Orius similis	■	■		■	Luo et.al., 2014
O. minutes	■	■		■	
Labidura riparia	■	■	■		Henneberry, 2007, Luo et.al., 2014
Chrysopa carnea		■			Henneberry, 2007
Adult *malachiids*		■			
Collops marginellus		■			
Notoxus calcaratus		■			
Nabis americoferus		■			
Nabis ferus,	■	■		■	Luo et.al., 2014
Paederus fuscipes		■	■		
Calosoma chinense		■	■		
Calosoma maximowiczi,		■	■		
Carabus coptobabrus,		■	■		
Isiocarabus fiduciaries,		■	■		
Chlaenius bioculatus,		■	■		
C. circumdatus,		■	■		
C. inops,		■	■		
Craspedonotus tibialis,		■	■		
Brachinus aeneicostis,		■	■		
Cicindela chinensis,		■	■		
C. elisae,		■	■		
C. sumatrensis,		■	■		
C. laetescripta,		■	■		
Chrysopa sinica,	■	■			
C. septempunctata,	■	■			
C. shansiensis,	■	■			
C. formosa,	■	■			
Hierodula saussurei		■			
Euborellia pallipes		■			
Apolygus lucorum	■	■			
Pardosa milvina			■		
Polybia ignobilis			■		
Spiders *	■	■			
Pipistrellus kuhlii (insectivorous bats)		■	■	■	Cohen et.al., 2020

Pink bollworm management – A global perspective

Management is one of the key issues. USA and Mexico relied on an area-wide approach using sterile insect release technique coupled with mating disruption, Bt-cotton, short-season and insecticide use for the eradication of the pest, in addition to the normally recommended IPM practices. China on the other hand deployed F2 hybrid seed of the single Bt gene Cry1Ac, which provided 25% non-Bt plants in the field for the management of pink bollworm, in addition to the commonly used IPM strategies. Pink bollworm is not a major concern in Australia and most countries in the African continent because of several factors, one of which is the enforcement of a closed season in Africa and the implementation ot IPM and IRM strategies in Australia. The synthetic pyrethroid group of insecticides are still used in Africa because of they are inexpensive and are recommended for a wide range of insects. Synthetic pyrethroids are still effective in controlling pink bollworms.

Turkey, Spain and Greece are a few of those countries that do not cultivate BT cotton and yet have successfully managed the pink bollworm through the use of pheromones and insecticides in addition to the cultivation of early maturing varieties and timely crop destruction. Uzbekistan is probably the only country that does not report the pink bollworm as a major pest of cotton. Turkey does not cultivate Bt or genetically modified cotton. Cotton is cultivated completely with irrigation, over an area of 518630 Ha and produces 977000-tonnes of fibre. Seeds are locally sourced; planting is done at a spacing of 70 cm X 10 cm with a plant population of 142857 plants per hectare and sowing to harvest takes between 5-7 months. Apart from the use of insecticides, cultivation of early maturing varieties, destruction of 'blind bolls' (ie bolls left after harvest) and use of sex pheromones for mass trapping have facilitated the management of the pest. Cotton is predominantly rotated with wheat in Turkey, where timely sowing of wheat necessitates harvest of cotton on time which by default enforces a 5-6 months closed season. Cotton cultivation in Turkey was expanded to newer areas with the availability of irrigation. The GAP area is a relatively new area of cotton cultivation in Turkey, where native natural enemies were reported to be abundant, thereby facilitating ecofriendly pest management.

Spain and Greece do not commercially cultivate Bt cotton although Bt cotton was tested in Spain (Garcia Olmeldo 2003). While half the seeds for sowing are imported in Spain, Greece uses locally available seed. Seeds are sown at a spacing of 90X10 cm and 90X6 cm with plant populations of 111,111 and 173,611 in Spain and Greece, respectively. Synthetic pyrethroid sprays based on trap catches targeting adult moths was the management tactic adopted by Spain (Duran et.al., 2000) while mass trapping and mating disruption appeared to be the method of choice in Greece. Thus, high density planting provided a quicker harvest of high yields thereby enabling a closed season which may have been responsible for the long-term suppression of PBW in these countries. The absence of PBW in Uzbekistan is interesting.

There are no clear explanations, but the possibility of PBW suppression due to repetitive innundative releases of *Trichogramma* egg parasitoids, for bollworm management over several years (Deguine et.al., 2008) cannot be ruled out.

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Seasonal Dynamics of Pink Bollworm in Cotton Ecosystems of India and Key Ecological Aspects for its Management

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Background

Originating from the Indo-Pakistan region (Saunders, 1843) and co-evolved with cotton (*Gossypium* sp, L.) as its host plant, the pink bollworm (PBW) *Pectinophora gossypiella* (Saunders), (Lepidoptera: Gelechidae), has become a pest of global significance to the cotton growing areas of the tropical and subtropical world (CABI, 2017). Generally, PBW is a late season pest, the infestation of which coincides with the onset of reproductive structures like squaring, flowering and boll development in cotton crop; accounting for colossal yield losses. The larvae of PBW feed on developing flowers buds and the seeds of green bolls of cotton plants, which causes rosetted damaged flowers; premature opening and heavy shedding of infested bolls; reduction in fibre length; and poor quality of lint due to staining (Singh et al., 1988). Being native to India, PBW had adapted to the wider climatic conditions of the country and is able to successfully complete its life cycle between 20°C and 35°C, with the optimal development temperature range between 25°C and 30°C (Peddu et al., 2020). The life cycle of PBW varies with prevailing temperatures and other environmental conditions during cotton growing season, being shortest (35-37 days) during the relatively warmer months of July to October, and longest (59-73 days) during the cooler winter months of November to January (Fand et al., 2021). During the post-cotton season (February to June), PBW larvae hibernate in leftover bolls on cotton stalks that are either standing in the field and or stacked on field bunds, and also in the infested seeds of cotton lint carried to the market yards and ginneries (Mallah et al., 2000; Kranthi, 2015).

Appearances of adult moths *vis-à-vis* commencement of reproductive phenophases of cotton are the two prerequisites critical to the successful field infestation of PBW. Given that these conditions are met, generally it takes about two weeks (14-16 days) from the date of beginning of moth emergence to manifestation of field symptoms of PBW damage in terms of rosette flowers and/ or green boll infestation (Fand et al., 2021) The sequence of events occurring between the moth emergence and manifestation of the symptoms of PBW damage in cotton field are depicted in Figure 1. Because of its cryptic habitat (entire larval development is completed inside the bolls) ensuring protection from insecticidal applications and natural enemies, PBW establishes and perpetuates more easily compared to other insect pests (Kranthi, 2015; Fand et al., 2019). Thus, once the larvae enter the bolls, exogenous insecticide applications often become futile resulting in control failures. Considering the preoviposition period of 2-3 days after moth emergence and the egg incubation period of 4-5 days, altogether, a very narrow window of 7-8 days is available for coincidence of the management actions with oviposition and egg hatching in PBW, with a pre-requisite that the timing of moth emergence is known. The population dynamics and critical biological events in the life cycle of PBW — such as moth emergence, oviposition, egg hatching and larval development — are contingent mainly upon the crop microclimate (temperature, relative humidity and soil moisture) and crop phenology (onset of squares, flowers and green bolls). In view of recent re-emergence of PBW due to resistance development against *Bacilus thuringiensis* (Bt) cotton, and its serious threat to cotton cultivation in India, understanding its seasonal dynamics is crucial in devising ecologically safer and economically sound pest management programmes for this notorious insect pest.

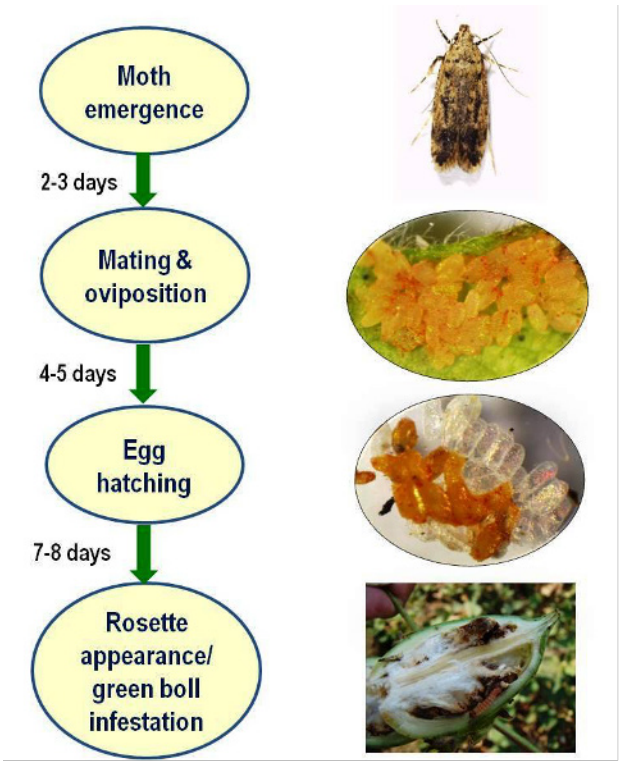

Figure 1. Sequence of events between moth emergence and manifestation of symptoms of PBW damage in cotton field (Adopted from Fand et al., 2021. *Scientific Reports,* 11(436).

Seasonal dynamics of PBW in cotton

Seasonal dynamics of PBW infestation *vis-a-vis* cotton phenology recorded during cotton growing season of 2019-20 using non-Bt cotton cultivar Suraj grown at experimental field of ICAR-CICR, Nagpur (India) is presented here as an example (Figure 2). The crop was completely unprotected from PBW infestation. The moth emergence from previous season's hibernating larvae started in mid-June (data not shown in graph); however it was not consistent as revealed by a very low number and discrete catches of male moths recorded at weekly intervals in pheromone traps installed along the field bunds and internal roads of experimental farm of ICAR-CICR, Nagpur. This irregular flush of moths that continue to emerge until July end to first week of August was supposed to be *suicidal emergence* as the cotton crop sown during the end of June to first week of July did not bear fruiting structures like squares and flowers for the emerging moths to oviposit and initiate the new season's infestation.

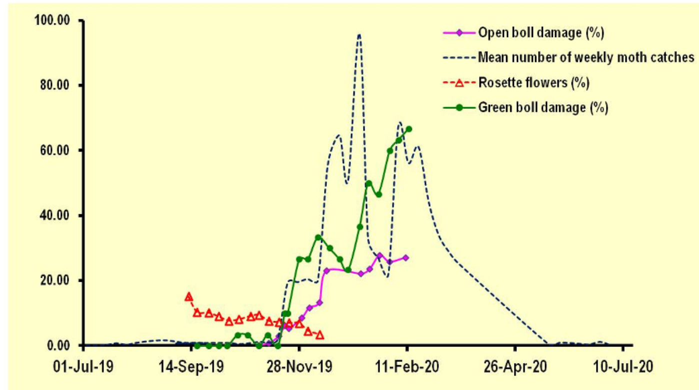

Figure 2. Seasonal dynamics of PBW infestation on cotton crop

Fairly low but consistent moth catches obtained from mid-to-late August seems to have established on the squares and buds giving rise to the rosette flowers from second week of September and subsequently the damage to green bolls from the second week of October. The flower infestation in terms of percentage of rosette was initially above the economic threshold level (ETL) (≥10%) through September, then remained closer to ETL (≤10%) until the end of October and declined steadily thereafter. This may be due to a shift in PBW preference from the squares and flower buds to the green bolls.

Staggered sowing of cotton crop spread over a span of one month due to delayed onset of monsoon at the beginning of season; continuous rains from August to October months resulting in prolongation of the vegetative growth phase; and delay in the flowering and boll formation are the primary reasons for the late beginning of PBW infestation in cotton during the cropping season of 2019/20 compared to earlier seasons (ICAR-CICR, 2020). The PBW infestation in terms of percentage of green boll damage was reasonably below ETL (≤10%) until mid-November, after which it started building up with its first seasonal peak that occurred during mid-December and the two subsequent peaks during mid-January and mid-February. A progressive increase in field infestation with the advancement of the crop season was observed. This was indicated by steep increase toward the end of the season in number of moths captured in pheromone traps and severity of damage to the green bolls and open bolls. For successful development of different life stages of PBW, the lower and upper threshold temperatures of 13.0°C and 34.0 °C and a thermal requirement of 500 ± 5 degree days (DD) are required (Fand et al., 2021). Based on mean DD accumulated between the consecutive moth peaks obtained in sex pheromone traps, it is evident that PBW completed four non-overlapping generations in a cropping season. Considering the intermittent peaks of moth catches, an additional three overlapping generations may be completed by the pest under favourable environmental conditions.

Fitting of linear regression equation (Y = a + bX, where 'Y' is the field infestation of PBW in terms of either rosette flowers, green boll damage or open boll damage; 'X' is the moth trap catch value two weeks prior to appearance of field damage; 'a' is the intercept and 'b' is the slope of regression equation) indicated that the data on mean rosette flowers and mean green boll damage were reasonably related, with the male moth trap catches recorded at two weeks prior to manifestation of damage symptoms in the cotton field. Similarly, the mean open boll damage was linked to corresponding green boll damage recorded two weeks prior to open boll damage (Table 1, Figure 3).

Table 1. Parameters of linear regression equation fitted to estimate the relationship between field symptoms of PBW infestation and moth catches in sex pheromone traps

S.N.	Parameter	Intercept (a)	Slope (b)	df	*F-stat*	*t-stat*	*p*	R^2
1.	Rosette flowers (%) vs moth catches	3.25 (0.95)	7.31 (0.98)	1,6	54.98	7.41	0.001	0.92
2.	Green boll damage (%) vs moth catches	16.44 (3.66)	0.31 (0.08)	1,9	15.01	3.87	0.002	0.67
3.	Open boll damage (%) vs green boll damage (%)	6.98 (2.02)	0.77 (0.14)	1,13	30.04	5.48	0.0001	0.71

*Figures in parentheses are standard errors

Key ecological aspects for devising management strategies for PBW

The squares, flower buds and developing green bolls of cotton plants are the preferred feeding sites for PBW. Normally, the first in-field generation of PBW is completed on squares and flowers, whereas the second generation onwards are completed on green bolls (Ellsworth et al., 2006). The moths emerging from previous season's overwintering population lay eggs on young floral buds (squares). The larvae feed and develop within squares leading to formation of rosette flowers and pupate either in rosette flowers or in soil debris at the base of plant. This sequence of events is repeated for subsequent generations on green bolls instead of squares and flowers (Sevacherian and El-Zik, 1983; Ellsworth et al., 2006). Thus, due to availability of flower buds and bolls, the cotton plant becomes a favourable host for PBW infestation from approximately 40-45 days after sowing (DAS), which usually correspond to late August to mid-September, when the mean environmental temperatures ranges between 24°C to 27°C. The generation developing on squares and flower buds generally takes 35-37 days to complete, by the time the crop will reach 70-80 DAS. With the ample availability of bolls, the second generation develops on green bolls from 80

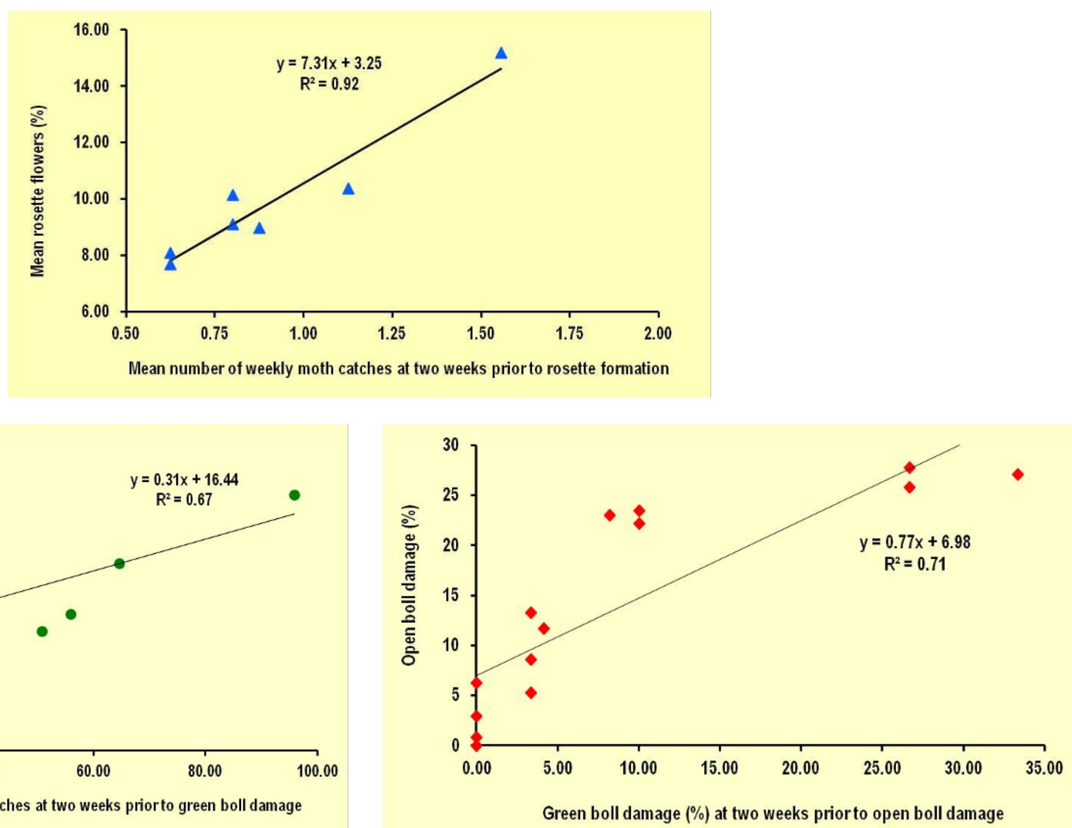

Figure 3. Relationship between field symptoms of PBW infestation and corresponding moth trap catches: rosette flowers (%) vs moth catches (a); green boll damage (%) *vs* moth catches (b); and open boll damage (%) vs green boll damage (%) (c).

to 100 DAS, whereas a third generation occurs when the crop reaches ≥120 DAS. Considering the low survival rate of PBW on squares relative to green bolls, a large population is not expected to build up during early periods of the cotton season. Time-series analysis of data on PBW moth trap catches across different locations of India revealed that the PBW population usually reaches its peak in third generation or later (Fand et al., 2021). This important ecological information is critical to the management of PBW. The management actions oriented toward the monitoring of pest activity (eg installation of pheromone traps) can be initiated during early periods of crop season in order to make timely pest management decisions. The maximum damaging population of PBW during the third generation and onward can be targeted to achieve effective control and to avoid economic yield loss to the cotton crop.


Photograph: Upender Mahesh

A graph of seasonal dynamics (Figure 2) indicated higher population of PBW built up beginning in mid-December. If the cotton crop is standing in the field for a prolonged duration beyond the normal recommended cropping window, the intensity of boll damage by PBW increases. However, in absence of cotton as its principal host, the population starts diminishing in preparation for overwintering either in the infested bolls of stalks or infested seeds of cotton (Mallah et al., 2000; Kranthi, 2015). Therefore, timely termination of the cotton crop either by the end of December through mid-January has been advocated as one of the important strategies of PBW management in India (Kranthi, 2015). Based on the degree day-based phenology prediction model, Fand et al (2021) have shown that at least two in-field generations of PBW could be prevented and yield losses can be minimised by adopting timely crop termination. The graph (Figure 2) also shows that moth catches started declining after mid-February, however they continued to emerge through April. From May to mid-June there was almost a complete absence of moth emergence. Taking into account the new flush of moths that are expected to emerge from mid-June with onset of monsoon showers, the pre-monsoon sowing (April-May) adopted in few scattered irrigated pockets is not advisable in order to escape the damage from suicidal population of PBW. Timely removal and destruction of infested crop residues to reduce the inoculum load of hibernating pests and monitoring and management of off-season flushes of moths emerging from infested cotton seeds through installation of sex pheromone traps and/ or light traps in the premises of market yards and ginneries are required.

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Biology of Pink bollworm, *Pectinophora gossypiella* (Saunders) (Lepidoptera: Gelichiidae)

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Introduction

The Pink bollworm, *Pectinophora gossypiella* (Saunders) (Lepidoptera: Gelichiidae) has recently emerged as a major problem in cotton production systems of central and south India. Though PBW was a major pest prior to 1980 in most parts of India, especially in north India, it is widely believed that the introduction of cotton-wheat rotation system in north India — which prompted the adoption of short-season cotton cultivars to fit in the crop rotation scheme — was mainly responsible for a decline in severity of pest infestation in north India. Synthetic pyrethroids were introduced in 1980 in India which had a strong control effect on PBW coupled with the newly introduced practice of timely termination of the crop to avoid late-season, PBW-damaged bolls appear to have relegated PBW into a minor pest in central and south India. The use of pyrethroids declined as American bollworm resistance levels to pyrethroids accelerated leading to a possible return of PBW when *Bt*-cotton was introduced in 2002 in India, which is believed to have kept PBW under check at least until 2010 when the pest developed resistance to *Bt* cotton.

PBW is a difficult pest to control because it is an endocarpic pest. However, with a good knowledge of its biology and ecology it will be possible to develop robust, eco-friendly management strategies that can keep the pest under check, as shown in USA, China and many other countries.

Bionomics of *P. gossypiella* on alternate host plants

Pink bollworm is a functional oligophagous pest with its main host range covered under Malvaceae. However, PBW also feeds on plants of a few other families. Cotton is the most preferred and major host for pink bollworm. In addition to cotton, it has a host range of other malvaceous plants such as *Hibiscus cardiophyllus* Gray, *Hibiscus coulteri* Gray, *Hibiscus denudatus* Gray. Two species of cultivated malvaceous plants were also recorded as host plants, namely, okra (*Hibiscus esculentus* L.), and hollyhock (*Althaea rosea*L.) by Rude (1932). Biology and morphometry of PBW on different host plants *viz*., okra (*Abelmoschus esculentus*) and Tuturbenda (*Abutilon indicum*) were evaluated (Table 1).

Biology

PBW completes its life cycle in 32-37 days. Female moths deposit eggs near calyx or bracts or near cotton bolls at the time of flowering. Under optimum conditions, neonates emerge after 3-5 days after oviposition and enter cotton bolls shortly after emergence. The larvae feed on developing seeds inside the boll. Larvae complete four instars inside bolls in 12-15 days and move out sometimes through an exit hole near the upper end of the boll to drop down and pupate in the top 5 cm of the soil. The pupal period lasts for 7-8 days. Adults exhibit nocturnal behaviour. Females are polyandrous. Oviposition occurs

Table 1. Biology and morphometrics of *P. gossypiella* on different host plants

S. No	Stage of the insect	Bt cotton ʌ=10 Mean (days) ± SD	Range (mm)	Non Bt cotton ʌ=10 Mean (days) ± SD	Range (mm)	Okra ʌ=10 Mean (days) ± SD	Range (mm)	Abutilon ʌ=10 Mean (days) ± SD	Range (mm)
1.	Incubation period	4.00± 0.00	4.00-4.00	4.00± 00	4.0-4.0	4± 00	4-4	4± 0.00	4-4.0
2.	I instar	5.20± 0.91	4.50-6.00	4.70± 0.48	4.5-5.0	4.5± 0.52	3.98-5.02	4.70± 0.48	4.5-5.0
a.	II instar	6.50± 0.52	6.50-7.50	5.50± 0.52	5.0-6.0	4.5± 0.66	3.84-5.16	5.35± 0.41	5.5-7.0
b.	III instar	7.0± 0.52	6.50-7.50	6.00± 0.33	5.67-6.33	5.25± 0.26	4.99-5.51	6.25± 0.26	6.00-6.50
c.	IV instar	6.25± 0.67	5.50-7.00	5.70± 0.94	4.5-6.0	4.5± 0.62	3.88-5.12	4.75± 0.92	4.5-7.0
d.	Total larval period	24.90± 1.42	22.8-26.92	21.9± 1.39	19.0-23.0	18.7± 1.40	17.35-20.1	20.85± 1.45	19.40-22.30
3.	Pre- pupa	1.55± 0.10	1.40-1.60	1.55± 0.10	1.40-1.60	1.55± 0.10	1.40-1.60	1.5± 0.00	1.50-1.50
4.	Pupal period	7.00± 0.80	6.20-7.80	7.25± 1.00	6.50-8.50	7.50± 1.00	6.5-8.5	7.25± 0.25	7.0-7.50
5.	Adult longevity								
	a. Male	8.90± 0.39	8.50-9.50	9.05± 0.15	8.90-9.50	9± 0.33	8.67-9.33	9± 0.33	8.50-9.50
	b. Female	9.50± 1.17	8.0-11.00	12± 1.17	10.83-13.17	9.5± 1.13	8.37-10.63	10.0± 0.81	9.19-10.81
6.	Pre oviposition period	2.5± 0.00	2.50-2.50	2.50± 0.00	2.5-2.5	2.5± 00	2.5-2.5	2.5± 0.00	2.5-2.50
7.	oviposition period	7.0± 0.00	7.0-7.00	7.5± 0.00	7.5-7.5	7.5± 00	7.5-7.5	7.5± 0.00	7.5-7.50
8.	Fecundity (no.)	128.5± 10.7	177.8-139.2	162± 10.60	151.9-173.1	124.5± 10.9	113.6-135.4	132.5± 10.5	122-143.0
9.	Total life cycle male	46.40± 1.60	45.19-49.06	43.75± 1.54	42.21-45.29	40.80± 1.21	39.2-43.3	42.85± 0.95	40.77-44.48
10.	Total life cycle female	47.0± 2.34	45.20-51.09	45.7± 2.26	42.3-49.5	41.30± 1.75	4 0.20-45.50	43.85± 1.60	41.30

(Source: Nagamandla Ramya Sri and Uma Maheshwari (2021)

within 1-2 days after mating. Each female lays 150 to 400 eggs depending on the number of matings. PBW normally completes four cycles in a cropping season, sometimes extending to a total of 6-7 cycles in a season depending on the crop duration.

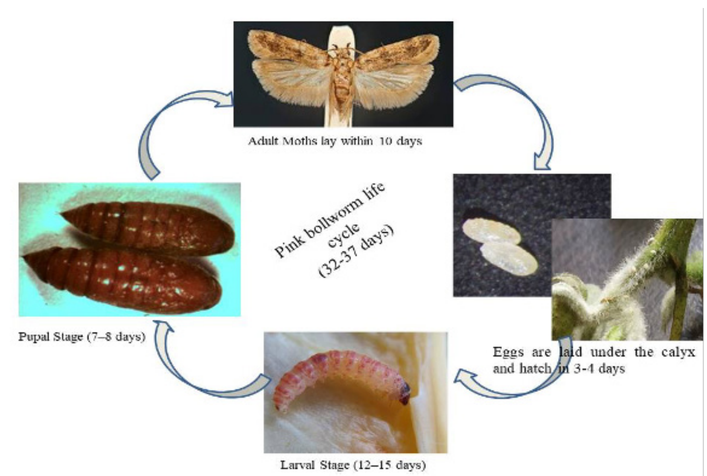

Figure 1. PBW life cycle

Eggs

Eggs are laid singly, or sometimes in small clumps of 2-3 eggs. Initially translucent white, eggs turn orange the next day. The first-generation moths lay eggs mostly on cotton squares inside the bracts. The second and the subsequent generation moths lay eggs mostly near the base of bolls behind bracts. Eggs hatch after 3-4 days of incubation. Studies show that moths do not show any specific ovipositional preference to Bt or non-Bt cotton.

Larvae

Soon after hatching, the neonates bore into squares or bolls. Larvae feed on the developing ovaries in squares as the square development continues. By the time the flower opens, it has been deformed into a rosette flower which shows twisted petals. Rosette flowers rarely succeed in normal anthesis, pollination and boll development. Studies show that moths resulting from larvae that feed on squares are less fertile compared to moths the are formed from larvae that feed on bolls. Larvae prefer bolls to squares or flowers. Larvae feed on tender seeds in developing bolls and devour 4-5 seeds to complete their feeding cycle in 4 instars. The whole locule in which the larva feeds generally rots due to secondary infection caused by pathogens that gain their way through the entry hole. Fibre in

Figure 2. PBW damaged Rosette flower, green boll and damaged loculi in open boll

the damaged locule does not develop properly and leads to a clear economic loss. Exit holes also cause damage of the entire loculi. The fourth instar larva enters into diapause under short day-light and dry conditions. Most diapausing larvae prefer to stay inside a seed or sometimes in a double-seed inside the boll, whereas most non-diapausing larvae exit out of the boll to pupate in plant debris and the upper layers of the soil.

Pupa

Pupae are reddish brown in colour. Pupal length ranges from 7-8 mm and breadth at 2.5-3.0 mm. Male and female pupae can be differentiated by the shorter distance on the dorsal side between genitalia and anal aperture in the male pupa compared to a longer distance in the female pupa.

Adult

Adult pink bollworm moths are mottled grey in colour. The moths are 7-8 mm long and 2.5 mm wide in a normal folded posture. On expansion the wingspan extends from 16 to 18 mm. Male moths mate 2-3 days after emergence while females take slightly longer. Males and females could mate more than once with different partners. Oviposition period extends up to 10 days. The moths feed on leaf nectar mostly. Moths are known to live for 1-2 months.

Figure 3. Pink Bollworm Adult

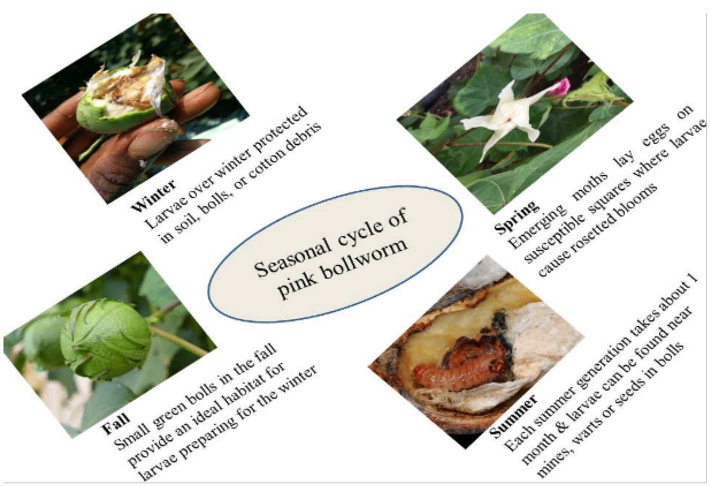

Figure 4. Pink Bollworm Seasonal Cycle

Diapause

The final instar larva undergoes diapause generally in the fourth of fifth generation when winter sets in India, Diapause is very common in north India as can be seen in double seeds in harvested cotton. Diapause has also been reported in south India. Studies show that as winter sets in with shorter days and a cool dry weather, fourth instar larvae enter diapause by spinning a loose silky cocoon. Diapause can happen in immature bolls or unopened bolls or stored seeds or in soil and crop debris on the soil. Diapause is known to continue from a few months to 2.5 years. Diapause gets broken with return of normal seasonal conditions that generally favour the survival. A few moths that emerge early even before the crop is sown and therefore in the absence of food, are termed as 'suicidal population'.

Management based on biology and ecology

Several management strategies have been recommended based on PBW biology and ecology (Kranthi, 2015). A few important strategies are listed below:

- Timely termination of the crop and ensuring a cotton-free closed season leads to a low carry-over population and thereby low pestilence
- Removal and destruction of the last batch of unharvested immature bolls can potentially destroy diapausing larvae thereby minimizing the pest inoculum of the subsequent season
- Area-wide pheromone-based mass trapping of the first-generation moths followed by mating confusion, light traps and insecticide application for the second-generation moths can effectively curtail pest populations from reaching ETLs in the subsequent stages of the crop.

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Reproductive Biology of Indian Population of Pink Bollworm, *Pectinophora gossypiella* (Saunders) (Lepidoptera: Gelechiidae)

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Introduction

Pink bollworm (*Pectinophora gossypiella*) (Saunders) has recently evolved as the most destructive insect pest in cotton in India. It is widely distributed all across the country wherever the crop is grown. The application of effective management practices for any pest largely depends on a thorough understanding of reproductive biology and its behaviour. Though, pink bollworm (PBW) is a key pest of cotton worldwide, information on reproductive biology is limited or poorly studied.

The study reported here was conducted at University of Agricultural Sciences, Bengaluru, and University of Agricultural Sciences, Raichur, India. Different behavioural parameters of *P. gossypiella* were observed in laboratory and field conditions.

Figure 1. Unmated spermatheca

Figure 2. Single mated spermatheca

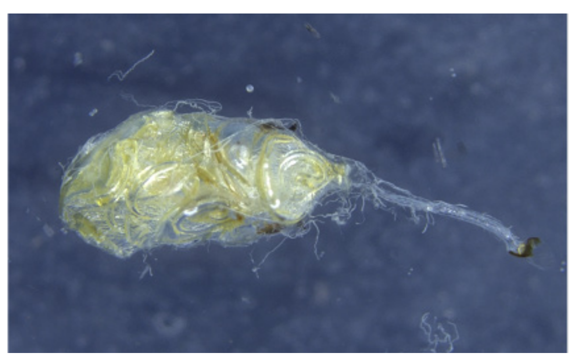

Figure 3. Multiple mated spermatheca

Emergence

Emergence of moths started from 11:00 hrs and continued up to 22:00 hrs and the peak moth emergence was observed between 16:00 to 18:00 hrs.

Flight

Flight activity of moths started after sunset (dark conditions), gradually decreased after 3:00 hrs and ceased completely after sunrise.

Mating

The majority of the moths initiated courtship behaviour two days after emergence, only in dark conditions. Mate finding behaviour was triggered by female calling behaviour and were characterised by rising abdomen, protrusion of genitalia and wing fluttering. Copulation started from 22:00 hrs and continued until 2:00 hrs, at a total mating duration ranging from 30 minutes to 2.45 hrs. The mean duration of mating was 1.12 ± 0.11 hrs. Female moths exhibit polyandry (mate with multiple males) and most of the times a female prefers to select a fresh male over spent males. A maximum number of six successful matings were observed and confirmed by dissecting the bursa copulatrix (Spermatheca) in female moth. The male moth is also polygamous in nature (mates with multiple females). A male moth can choose to mate with four females and re-mates with same female generally under a 'no choice' condition.

Oviposition

Majority of females oviposit during dark conditions, but a few moths were also observed to oviposit during the day. The females laid eggs in batches every time after successful mating. The fecundity of a female increases when it mates multiple times. A single mated female laid fewer number of eggs (82 ± 14) whereas a multiple mated female laid a copious number of eggs (216 ± 25). The length of ovipositional period varied from 4-11 days.

Reproductive fitness and life span

Female moths increase their reproductive fitness by multiple mating which paradoxically is negatively correlated with their life span. Moths with a higher reproductive fitness have a shorter life span. Thus, there is a trade-off between reproductive success and adult life span for both males and females. An unmated female lives longer (20 ±3 days) compared to single mated (16 ±2 days) and multiple mated females (12± 2 days).

Reproductive fitness and light traps

The study attempted to examine the proportion of males and females (both mated and unmated) that were attracted to a light trap in cropping seasons over two years. Light traps attracted a larger number of male moths over females. Amongst the females captured, single mated females were significantly more and very few multiple mated females were attracted. Installation of light trap in the cotton field could also be considered as an important management tool to reduce PBW oviposition.

Management strategies for PBW in Indian conditions

Replace cotton hybrids with short duration varieties

In India, farmers depend on hybrids for high yields. Unfortunately, hybrids continuously produce flowers and bolls for more than 180 days which creates ideal conditions for PBW to infest throughout the cropping period. There is a need to switch over to short duration varieties, where boll formation is completed within 120 days.

Varieties without pubescence

Unlike other insects, PBW female moths lay eggs without any secretion on the egg surface; hence it prefers tender parts (shoots and leaves) where pubescence is present. In glabrous varieties (devoid of pubescence), eggs do not attach properly to the plant surface and eventually fertility gets reduced.

Avoid mono-cropping

Growing cotton after cotton in subsequent seasons is advantageous for monophagous or oligophagous insect pests to survive and reinfest the crop year after year. Hence, farmers should grow different types of crops in rotation over year to ensure that malvaceous crops are not repeated over years. This will break the cyclic occurrence of PBW populations.

Installation of light trap

Even a small number of two to three light traps per hectare have the potential to attract a large number of males and females which has the propensity to reduce the population build up in the field.

Biocontrol

Release biocontrol agents like *Trichogramma bactrae* at two intervals, ie, flowering and boll formation stage.

Insecticides

Spraying insecticide with ovicidal property at flowering stage and boll formation stage can greatly help in reducing pestilence.

Can we disrupt PBW reproduction?

Both male and female moths mate multiple times and lay large number of eggs by increasing their reproductive fitness. There is a huge scope to develop products which could affect reproduction and reduce the egg load significantly.

Cotton Gin Trash Treatment System to Destroy and Prevent Carryover of Pink Bollworm from Ginning Mills

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Introduction

Cotton is known to harbour several pests. Among them Pink bollworm (PBW) is of serious concern. The pink bollworm (*Pectinophora gossypiella*) is a dreaded pest on cotton and is distributed throughout the world's cotton-growing areas. After the introduction of *Bt*-cotton in 2002 in India, there was an excellent response as manifested by low incidence of all bollworms because the *Bt* gene provided built-in protection against them. Since 2015, frequent outbreaks of PBW have been documented in the states of Maharashtra, Andhra Pradesh, Gujrat and Telangana because of the development of resistance to PBW in *Bt*-cotton (Kranthi, 2015). The rough estimates of incidence varied from 20%-60% resulting in estimated yield losses of 10%-30% but much more in quality impairment (Mayee *et. al.* 2018). PBW adversely affects ginning percentage and spinning quality of mature bolls. Pink bollworm infestation causes significant yield losses and affects quality of cotton fibre and thus adversely affecting income of cotton farmers.

Timely termination of *Bt*-cotton crop is an important strategy to break the seasonal cycle of PBW resulting in very low carryover of PBW populations and low PBW damage in the subsequent season. Diapausing PBW larva can survive in immature unharvested cotton bolls on stalks stored by farmers (Vennila et. al. 2007). Diapausing PBW larvae survive inside cottonseeds and gin trashes stored in cotton gins. Longer the storage, higher the carryover potential. Appropriate treatment needs to be given to gin trash before its disposal to break the seasonal pest cycle.

Concern of Pink Bollworm residual infestation from Ginning Industry

Concern is raised in India that cotton ginning industries serve as sites of inoculum of PBW. In India cotton fields are small and the gins are located at considerable distances from one another, which leads to the transport of PBW over long distances. This could result in the dispersal of PBW in the neighbourhood locality where cotton is ginned. Transportation of seed cotton from infested area or the gin to the non-infested gin leads to the rapid spread of PBW. It is important to prevent the spread of PBW through ginning industry. During ginning operation there is potential for PBW to survive and pass-through pre-cleaning and ginning machinery which gets segregated into cottonseed, lint and gin trash (Hughs et. al., 1994). Ginneries are equally responsible for the spread of PBW through gin trash which has not been commonly acknowledged.

Necessity of Mechanical Treatment to Gin Trash before Disposal

Destruction of PBW larvae and pupae in gin trash and gin waste in ginning industries can effectively curb the spread of PBW from ginning mills. Unless measures are taken to control and eradicate PBW in ginning industry, its menace is likely to continue. Mostly live PBW find its way into the gin trash from all cleaning machinery, cyclone wastes and immature and unopened bolls. Currently in ginning industry gin trash is scattered all across the industry premises, from where diapausing larvae survive and PBW moths emerge and spread to neighbouring areas causing pestilence in the subsequent seasons. Therefore, it is essential to crush all gin trash mechanically with specific devices or systems in such a way as to cause destruction of all the stages of the pest. After that has been done it is safe to dispose of the treated gin trash.

Mechanical Gin Trash Treatment System

A gin trash treatment system (Fig.1) comprising of centrifugal trash fan, cyclone and compactor was developed with a capacity of 2.5 tonnes of trash per hour with an aim to crush and treat gin trash mechanically in such a way that all PBWs are destroyed. A centrifugal trash fan was developed to treat gin trash prior to its disposal. The action of the fan kills the pink bollworms in the trash, when gin trash is passed through the impeller of a trash fan operating according to pink bollworm quarantine regulations laid down by USDA. Centrifugal trash fan was developed as per the USDA regulations for wheel diameter, inlet size, and speed of fans for treating gin trash. Cyclone (1D-3D) was developed as an air pollution abatement system on high pressure centrifugal fan discharges, to separate air and waste in cotton gins. A compactor was developed for volume reduction of gin trash after treatment. The compaction enables cost effective disposal of the treated gin trash. A gin trash treatment system has been commercialised for commercial scale production and marketing.

Figure 1: Cotton gin trash treatment system

Protocol to Destroy PBW in Gin Trash

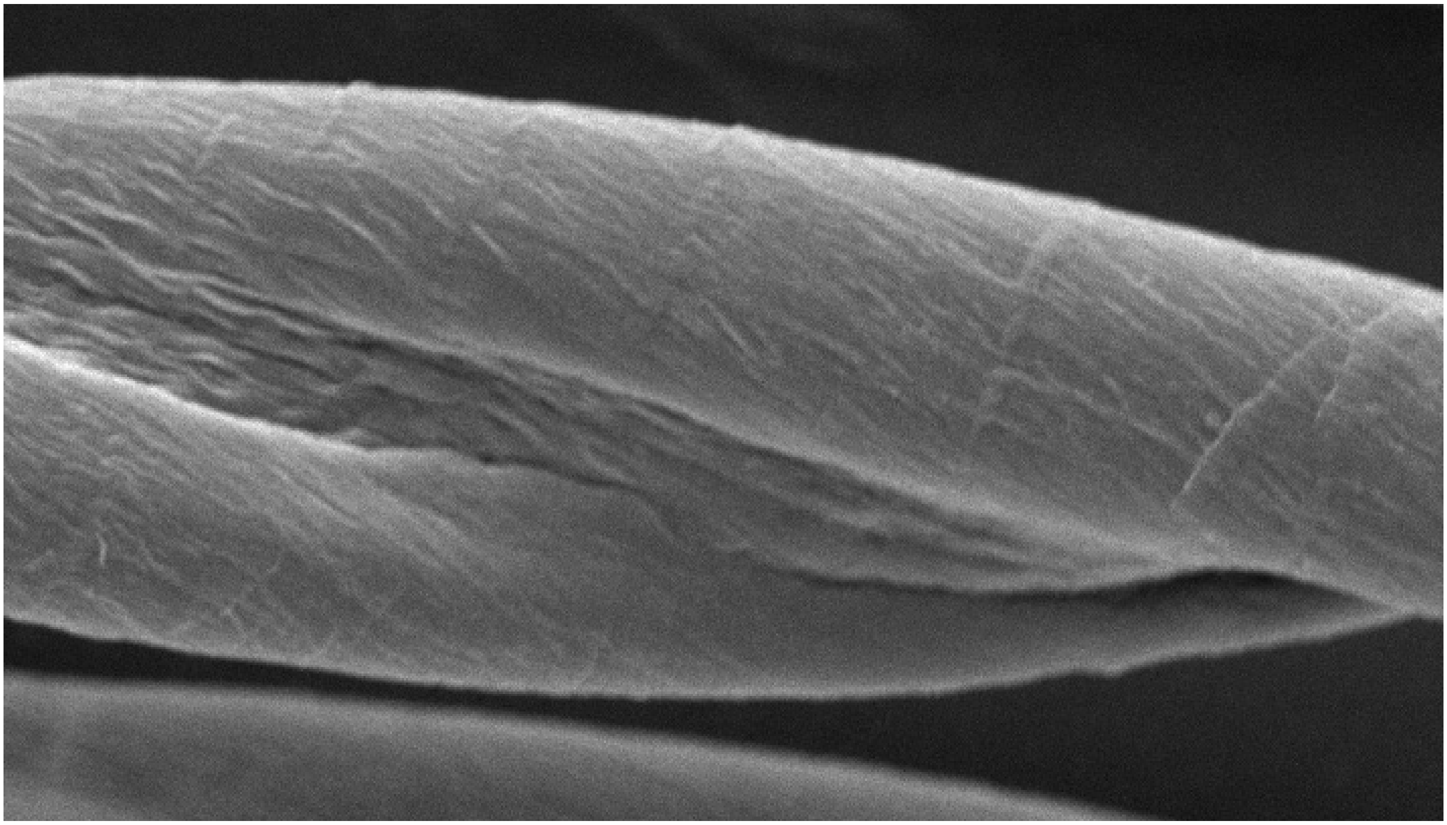
A process protocol has been established to destroy pink bollworms from cotton gin trash by using gin trash treatment system. The trash fan wheel which crushes gin trash should have a minimum of six straight blades with minimum fan diameter of 490 mm. The trash fan should generate an air volume of 4800 m^3/h and run at a pressure of 363 mm wgp. A minimum fan tip speed of 4192 m/min i.e., about 3000 rpm should be maintained. The minimum air velocity through 254 mm ducting should be more than 17 m/s. The 1D-3D cyclone with diameter of 815 mm and height of 2445 mm needs to be employed to separate out air and trash passing through the trash fan with a pressure drop of 363 mm wgp. The screw conveyor of the compactor should have a pitch and diameter of 320 mm and should run at a speed of 72 rpm. With this protocol the developed system attains the intended objective of destroying PBW with 100% mortality rate of larvae and pupae.

Effect on Pink Bollworm Infestation of Ginning Performance

Ginning performance of PBW-infested cotton compared to un-infested cotton on DR gin showed 17%, 14%, 25% and 11% reduction in ginning percentage, fibre length, uniformity index and tenacity respectively whereas short fibre content and degree of yellowness increased by 67% and 42% respectively which is undesirable. Deterioration in colour grade of cotton of PBW infested cotton is observed to deteriorate from middling (31-1) to strict low middling (41-1) compared to un-infested cotton as per HVI colour grade chart for American upland cotton (USDA). Scanning Electron Microscope (SEM) analysis indicated poor fibre development and growth of microbes on PBW infested cotton in comparison to healthy cotton (Fig.2). The deterioration in ginning percentage and fibre quality has significant impact on the economic value of cotton thus affecting cotton producers and processors. (Fig. 2)

Suggested Measures for the Indian Ginning Industry

A gin trash treatment system along with best management practices comprising of regulatory and legislative measures are suggested to destroy and prevent dissemination of PBW through gin trash from Indian ginning industries to the



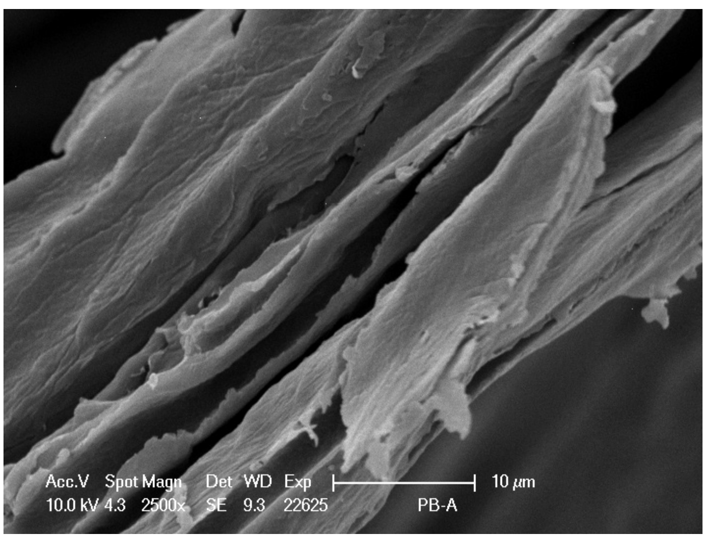


Figure 2: SEM images of healthy (above) and pink bollworm infested cotton fibre (below)

neighbourhood cotton fields. For successful elimination of PBW, the installation of a gin trash treatment system should be made mandatory in each ginning industry in India. Government regulation needs to be issued for mandatory use of this technology by the ginning industries. To encourage ginners to instal the developed system — incentives in terms of subsidy or additional cost compensation per bale ginning and pressing charges for ginners installing the system — need to be provided. Best post-harvest management practices based on cultural, mechanical and chemical control measures need to be promoted in the ginning industry. These measures would put cotton producers in a position to grow cotton profitably by increasing yields, reducing chemical inputs and preserving fibre quality.

Pink bollworm larvae undergo diapause in the last instar larval stage and constitute a significant number of populations that rest for several months and emerge only under favourable conditions to cause severe damage to cotton in the subsequent seasons. The diapausing larvae undergo into their resiting stage inside seeds or by weaving double seeds or inside unharvested immature bolls. Destroying the immature bolls or the seeds harbouring diapausing larvae is an important stragegy to control pink bollworms in India.

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Pink Bollworm Management in West Africa

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Abstract

The pink bollworm (PBW) called *Pectinophora gossypiella* is a serious pest of cotton in West Africa, especially in coastal countries like Benin, Côte d'Ivoire and Togo. Management of this insect is essentially based on knowledge of its emergence, seasonal occurrence and outbreak patterns, use of sex pheromone traps, and adoption of cultural and chemical control methods discussed in this article. Following the observed decreases in the sensitivity of *P. gossypiella* to Cypermethrin and Deltamethrin, studies in search of alternatives to pyrethroids are underway to prevent cases of resistance of this endocarpic caterpillar.

Keywords: *Cotton*, *Pectinophora gossypiella, Management, West Africa*

Introduction

The pink bollworm, also called *Pectinophora gossypiella* (Saunders, 1844), is one of the most destructive pests of cotton in tropical and subtropical regions of the world (Ingram, 1994). The current geographical distribution of *P. gossypiella* is closely linked to that of cultivated *Gossypium*. This area has expanded considerably with the establishment or extension of cotton cultivation throughout the world and in direct connection with intercontinental exchanges carried out in this context (Le Gall, 1995). In West Africa, it is found in humid zones where rainfall is about 1000 to 1500 mm per year. This corresponds to levels recorded in central Benin, Togo and Côte d'Ivoire (Ton, 2004). Resistance of the African bollworm *Helicoverpa armigera* to pyrethroids with which *P. gossypiella* shares the same habitat has prompted the need to search for alternatives to pyrethroids. The observed decrease in the sensitivity of *P. gossypiella* to Cypermethrin and Deltamethrin (Ochou *et al*., 2018) and the fact that it lives within the cotton boll makes it difficult to manage PBW. These situations have led to the adoption of integrated pest management strategies to limit its damage. The IPM strategies include:

- knowledge of the factors that influence PBW population densities,
- Use of pheromone traps,
- Cultural control, and
- Chemical control.

Methodology

The use of pheromone traps followed by a weekly observation of caterpillars in the field helped in the identification of appearance and outbreak periods of the pest. The proposed cultural control methods took into account the insect's bio-ecology. Efficiency studies of active ingredients against several species of pests including *P. gossypiella* have been carried out in different member states of the Regional Program for Integrated Cotton Production (PR-PICA) and summarised in a poster (PR-PICA, 2017). The list is not exhaustive.

Integrated Management of *P. gossypiella* Populations

Knowledge of the population dynamics of *P. gossypiella*

Monitoring of the seasonal dynamics of adult *P. gossypiella* populations in pheromone traps and larvae in fields within the sub region revealed that adult *P. gossypiella* populations were present almost throughout the season and even in-between the seasons, in low densities. Adult populations were few in number due to unfavourable environmental conditions from January to April. The caterpillars appear in cotton fields between late August and early September. The largest peak of caterpillar population was observed between September and December. Weather changes are not very favourable to the development of the species during other months. The optimal protection period against this pest will be from August ending to December, to take into account the first generations (Germain *et al*., 2018; Gustave *et al*., 2019).

Pheromone traps

The use of sex pheromone traps (funnel traps or glue traps) can be used to control this pest by capturing adult males or monitoring the evolution of its population (Boguslawski *et al*., 2001; Germain *et al*., 2018). A locally made pheromone trap, made from an empty mineral water bottle of 1.5 litres was used to monitor *P. gossypiella* populations. Detection of the pest using this sex pheromone trap, supplemented by weekly visual observations in the field, allowed preventive measures to be taken and integrated control methods to be implemented (Germain *et al*., 2018).

Cultural control

Cultural control methods play a key role in managing the populations of *P. gossypiella* (Vennila *et al.*, 2007). Cultural control strategies involve manipulation of the pest habitat to reduce its populations. There are a range of farming practices to be aware of: site selection, crop rotation, choice of sowing and harvest dates, tillage (ploughing), destruction of plant residues (just after harvest), use of trap plants, cover crops (legumes), mixed cropping and weeding. Because PBW populations are conditioned mostly by weather factors to occur at particular time periods during the season, an intelligent choice of sowing period can be determined to prevent the coincidence of larval occurrences and outbreaks with the peak boll formation stage. Sowings carried out from June 21 to 30 (4^{th} decade) helped to significantly reduce the population density of *P. gossypiella* in Benin (CRA-CF, 2018). Vennila *et al.* (2007), proposed that seeds should be dried for 6-8 hours in the sun and that seeds should be systematically de-linted before transport to prevent the spread of the pest. Uprooting of cotton stalks just after harvest and destruction of immature unharvested bolls (by crushing) stops the cycle of pest development.

Chemical control

Synthetic pyrethroids have been effective in controlling PBW infestation and are considered as reference products used for the control of *P. gossypiella* for over the past two decades. Combinations of pyrethroids with Spirodiclofen as basic ingredients have been effective (PR-PICA, 2017). A botanical insecticide produced from neem was found to be an effective alternative to synthetic pyrethroid insecticides such as cypermethrin and deltamethrin, to which PBW has been showing decreased susceptibility in recent years (Bonni *et al.* 2018). Protection programmes based on the mixture of neem aqueous extract and emamectin-acetamiprid formulations showed better control of PBW that cohabits with *H. armigera* (Bonni et *al.* 2018).

Conclusion

Taking into account the period of appearance and outbreaks associated with crop residues, cultural (management of cotton stalks) and chemical control methods and the use of alternative active ingredients to pyrethroids, allow a significant reduction in the populations of *P. gossypiella*.

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Eco-friendly insecticides to manage pink bollworm, *Pectinophora gossypiella* (Saunders)

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Abstract

The pink bollworm, *Pectinophora gossypiella* is considered one of the main pests in Egypt. This pest has acquired resistance to many conventional insecticides, so it was necessary to identify new insecticides against the pink bollworm. The new insecticides are not only effective against this pest but are also less toxic to nontarget organisms and cause less contamination to the environment. These eco-friendly insecticides include bioinsecticides (bacterial insecticides, viral insecticides, fungal insecticides, secondary metabolites of microorganisms and pheromones), plant extracts and natural nanoparticles.

Key words: The pink bollworm, eco-friendly insecticides, environmental contamination

What are eco-friendly insecticides?

Eco-friendly insecticides can be defined as insecticides that are toxic for pests but have little or no effect on mammals and beneficial insects. This form of pest control is a healthier alternative to the toxic chemicals that are commonly used as insecticides; it protects humans and beneficial insects from risky side effects, while saving the environment from further damage as well. These insecticides are made with ingredients that are safer for beneficial insects and are just as effective on harmful pests as are many conventional insecticides.

Using of eco-friendly insecticides against the pink bollworm

Due to the hazardous nature of insecticides, environment contamination and the potential human risks, there has been an intensive interest in the search for eco-friendly insecticides that can be used as alternative agents against the major insect pests such as the pink bollworm. The eco-friendly insecticides include bioinsecticides, plant extracts and natural nanoparticles.

Using of bioinsecticides against the pink bollworm.

Bioinsecticides include microbial insecticides (bacterial insecticides, viral insecticides, fungal insecticides, secondary metabolites of microorganisms). Reyaz *et al*. (2021) isolated a novel indigenous *Bacillus thuringiensis* isolate, T26, which showed spores and crystals. This isolate was effective against the pink bollworm. This isolate was not only toxic to the pink bollworm but also exhibited a potential for the possible presence of novel factors responsible for its virulence which could provide useful tools for the insect resistance management in pink bollworm. Moustafa *et al*. (2019) used *Metarhizium anisopliae* and *Paecilomyces lilacinus* (fungal insecticides) against the newly hatched larvae of pink bollworm. Radwan *et al*. (2018) used Nucleopolyhedrovirus (NPV) against neonate larvae of the pink bollworm. Spinosad (secondary metabolites of bacteria) also was used against the pink bollworm. Sabry *et al*. (2014) used spinetoram (secondary metabolites of bacteria *Saccharopolyspora spinosa*) against the first instar larvae of the pink bollworm. The LC_{50} was 19 ppm. Spinetoram is more toxic against the pests than spinosad and also a systemic insecticide which is effective against the sucking pests and the larvae of the pink bollworm. Spinosad is not only effective against the pink bollworm but is also relatively safer to the natural enemies such as green lacewing, *Chrysoperla carnea*, seven-spotted ladybug, *Coccinella septempunctata* and *Trichogramma* wasps, *Trichogramma evanescens* (Sabry *et al.*, 2014). Hosny (1988) used synthetic insect pheromone gossyplure against the pink bollworm infestation. Three applications of Gossyplure reduced the pink bollworm population significantly. The obtained results found that gossyplure was more effective against pink bollworm infestation than the conventional insecticides.

Use of plant extract against the pink bollworm

Aqueous ethanolic extracts of two plants, *Calotropis procera* and *Ocimum sanctum* were used against pink bollworm larvae (Yousef *et al.* 2016). Rajput et al. (2017) evaluated three plant extracts against the pink bollworm, tobacco extract, neem extract and datura extract. Results showed that tobacco extract was the most effective followed by neem and datura extracts. Farooq *et al*. (2020) used a mixture of both bioinsecticide and plant extract; entomopathogenic fungi (EPFs) viz. *Verticillium lecanii*, *Metarhizium anisopliae* and *Beauveria bassiana* with *Azadirachta indica* extract alone and in combination against the larvae of pink bollworm. The results showed that the entomopathogenic fungi have a synergistic action with *A. indica* against the larvae of the pink bollworm.

Photograph: Upender Mahesh

Using of natural nanoparticles against the pink bollworm

Natural nanoparticles are natural materials converted into nano size such as silica and zinc oxide nanoparticles. Derbalah et al. (2014) used silica nanoparticles and zinc oxide nanoparticles against the first instar larvae of the pink bollworm. The results showed that zinc oxide and silica nanoparticles were promising natural materials against the pink bollworm. Saadiya and Abdelaal (2020) used ginger extract nanoparticles against the eggs and larvae of the pink bollworm. The results showed high efficacy against both eggs and larvae compared to conventional insecticides.

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Bt or no *Bt*, Integrated Pest Management Should be an Integral Part of Pink Bollworm Management on Cotton

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Dr KS Mohan's entire career of >40 years has been in the area of R&D of insect-control technologies for plantation crops, horticultural crops, cotton and corn. He was involved in stewardship of *Bt* cotton and *Bt* maize in India. Dr Mohan strategized and implemented IRM for Bollgard® and Bollgard II® through partnership projects with public & private stakeholders. He established and managed *Bt* trait quality assurance testing platforms on molecular events, season-long transgene(s) expression, bollworm-control efficacy, *Bt* resistance monitoring and product trouble shooting. Dr Mohan formulated IRM strategy for *Bt* maize through partnership projects on risk assessment. He is passionate about scientific outreach and public advocacy in biotechnology. Currently Dr Mohan provides consultancy in developing transgenic cotton for insect and virus control.

Introduction

The pink bollworm (*Pectinophora gossypiella*) (PBW) is a destructive bollworm of cotton severely affecting the quality of cotton lint with presence in all cotton growing countries and a difficult pest to manage. PBW has been an invasive pest into most countries and is thought to have originated somewhere in Southeast Asia and gradually spread to the cotton fields of the southwestern USA through the middle east carried through 'fuzzy' cotton seeds infested with the resting stages of the pest (Naranjo et al., 2002). In India, PBW infestation of cotton has been recorded by entomologists in early 1900 and was so severe in south India during the British rule that a legislation was enacted in the presidency of Madras on the movement of cotton seeds from areas with PBW infested cotton (Ramachandra Rao, 1921). PBW continued to be key pest on hybrid cotton which was developed in India using the American tetraploid *Gossypium* species. Though it is a global pest, much of the content in this chapter is directed to the situation in India because it is the only country where PBW populations have evolved field-resistance to *Bt* cotton since 2010. The problem in India is quite acute in view of the large acreage of *Bt* cotton (~11.5 million acres) available for the *Bt*-resistant PBW for multiplication making India a fit case for the re-visit of Integrated Pest Management (IPM) methods for PBW management.

PBW is typically a late-season pest of cotton in India, but if the carryover population from the previous season is large enough, as is the current situation in many states, then this pest appears early enough during the initial bloom period. Among the key Lepidopteran pests of cotton, PBW is difficult to control with insecticides because of its reclusive feeding habits within the developing cotton bolls. In contrast other caterpillar pests of cotton spend a considerable portion of their time feeding on the plant surface, and hence are within the reach of insecticides. The larval stages of PBW spend their entire period within the boll, feeding on the developing seeds. Very often, the grownup larvae hibernate with the scooped-out cotton seeds to tide over the unfavourable winter season. Many grownup larvae exit through a hole made in the boll and fall to the soil where they hibernate in the cracks in soil or in the plant debris in a loosely woven silk bag. Hibernation in seeds or soil is the pest's way of tiding over unfavourable environmental conditions when no cotton crop is available for the progeny to feed and grow. When the temperature warms up and with increasing daylight, the hibernating larvae quickly complete the life cycle and emerge into the open as moths. Emergence coincides with the availability of flowers and new bolls in the new crop.

Bt-cotton (single and dual *Bt*-gene versions) had effectively managed PBW in the USA and India because of the high sensitivity of the larvae to *Bt* toxins expressed by transgenic *Bt* cotton. However, the *Bt* cotton technology in India stood eroded with time because of evolution of field resistance to first Cry1Ac and then to Cry2Ab2, sequentially, in 2010 and 2016 (Tabashnik and Carrière, 2019). Indian cotton farmers did not plant the 20% structured refuge with *Bt* cotton, nor did they adopt the recommended Integrated Pest Management (IPM) practices along with *Bt* cotton. Both these measures, had they been adopted, could have delayed the *Bt* resistance evolution in PBW.

IPM: the forgotten strategy to manage pests in cotton cultivation

Globally cotton happens to be infested by a variety of bollworms and sap-sucking pests and thus is a fit crop for the application of IPM methods for pest management. The severity of pest incidence can be appreciated from the fact that prior to the *Bt* cotton era, ~ 45% of the total insecticides used in crop-protection in India was on cotton. Among the bollworms, the PBW is a problematic and an enigmatic pest. It is unique in the fact that PBW can multiply productively only on cotton -thus is functionally monophagous. Several malvaceous weeds have been recorded as hosts of PBW, but they are not productive; relative to cotton, okra is a good host but the total acreage relative to cotton is very small. The narrow-host preference had hastened resistance evolution to *Bt* toxins expressed by dual *Bt*-gene cotton in the absence of structured refuge or natural refuge in the form of diploid native non-*Bt* cotton varieties. *Bt* cotton with cultural practices to disrupt the pest-cycle would have managed the bollworms and the bio-control agents would have taken care of the sucking pests.

In the absence of any other disruptive technologies in the near future for the management of *Bt* resistant PBW, the only option available to the Indian cotton farmers is to rigorously adopt IPM measures as part of area-wide management aimed at disrupting the pest cycle. Fortunately, PBW can be managed with a simple to adopt cultural and cultivation practices directed at minimising the carryover of PBW population between cotton seasons and need to be dovetailed into IPM modules designed to take care of the entire pest-complex, all Lepidopterans and sap-sucking pests, inclusive.

Figure 1. Pheromone trap catch: 1035 moths in a single trap, Janthmer, Bhavnagar 2014

Management of *Bt*-resistant PBW on *Bt* cotton in India

Certain practices in cotton cultivation play a key role in managing PBW (Kranthi, 2015; Mohan, 2017) and these could be divided into post-harvest, off-season and pre-planting periods.

Deep ploughing

Soon after the last pick of cotton, ensure deep ploughing of cotton fields. This operation would not only destroy hibernating larvae of PBW in the cracks in the soil but also pupae of other bollworms like *Helicoverpa armigera*, *Spodoptera litura* and *Earias* spp. This is an important, but often overlooked, method to minimize PBW and other bollworm moths emerging from the soil.

Destroy residual immature bolls

Detach all unopened/improperly opened bolls from the cotton stalks and destroy them by burying/burning, because these bolls often contain hibernating PBW larvae in the seeds within the bolls. Extension bulletins often recommend destroying the cotton stalks using a rotavator. This is not necessary if the stalks are picked clean of the unopened bolls because cotton stalks form an important source of firewood in the villages. Sun-drying of unopened/improperly opened bolls is practiced in many parts so as to recover as much cotton fibre as possible but should be strongly discouraged.

Destroy gin waste and trash

Trash consisting of PBW-infested cotton seeds in the ginning mills form an important source of spread of PBW because harvested cotton comes from faraway places for ginning. It is a common sight to see heaps of trashed cotton seeds in cotton gins, many infested with PBW larvae/pupae. In addition to destroying such trash, the gins should install pheromone traps in the vicinity of the gin to trap all emerging PBW moths. Similarly, transport of PBW-infested cotton seed for oil extraction to adjoining districts/states should be discouraged through legislation, if possible.

Timely crop termination

New sowings of cotton should ideally consist of medium-maturity *Bt* cotton hybrids, especially in areas where PBW is endemic. Long-duration cotton crop gives ample opportunity for the PBW population to multiply further by another 3 to 5 generations after 120 days of the crop, resulting in a large carryover to the next season. If suitable short/medium maturity cotton hybrids are not available, the farmers can go for long-duration cotton, but the crop needs to be terminated after the third pick/ 120 days.

Avoid ratooning and prevent re-flush

Ratoon-cropping or 're-flush' of cotton is practiced in irrigated areas after the last pick and this practice fetches the cotton farmer some additional income by extending the cotton crop till 250 days. Re-flush cropping becomes widespread particularly when the first few pickings are impacted due to vagaries of climate. Typically, such areas have large endemic population of PBW, built over several seasons. Thus, re-flush in cotton should be a strict NO in such areas.

Pheromone-trap monitoring

In areas with large carryover of PBW from the previous season, farmers can expect early appearance of PBW infestation in flowers and squares at 60 days. PBW pheromone traps (@ 3 to 5/ha) should be used to determine if PBW population level has reached Economic threshold level (≥ 8 moths per trap on three successive days), to decide on insecticide application, as per the advisory from the local agricultural agency. If the PBW population is large, use more pheromone traps (@ 20 traps/ha) for the purpose of pest suppression through mating disruption.

Sampling and scouting

Regular scouting for PBW infestation in flowers or bolls can help assess the infestation level and help decide on the course of mitigatory action.

Figure 2. Collection of boll samples for monitoring

Figure 3. Examining harvested cotton for diapausing larvae

Figure 4. Bioassays for resistance monitoring

It is important to remember that IPM — and specifically the component of cultural and cultivation practices to break the pest-cycle of PBW — can be better achieved through wide-area participation. It will not help if only a few farmers adopt IPM measures.

In contrast, PBW in the southwestern US has been managed judiciously by the combination of high level of compliance on structured planting and the synergy between the efficacy of *Bt* cotton and an assiduously implemented program of release sterile PBW moths leading to a formal declaration that PBW has been eradicated from the PBW-endemic areas of south-western USA (USDA, 2018; Tabashnik et al., 2021). Can this success be replicated in India? It appears to be very daunting, primarily because of the lack of efficacy of *Bt* cotton in managing PBW because of *Bt* resistance issues and the huge investment needed to setup PBW breeding labs for mass-production, sterilization and release of radiation-sterilized moths. Till we have a viable new technology to manage the *Bt* resistant PBW populations, we have no other choice but to strength the IPM approach towards managing PBW on cotton. Another hard lesson learnt is: Whatever new technologies might come, they have to be used in conjunction with IPM methods.

Conclusion

The high-level efficacy of *Bt* cotton in managing the pink bollworm in the initial years of cultivation in India had made cotton farmers slack on two important fronts: planting of structured refuge and adoption of IPM practices. Evolution has been extracting its toll in the form *Bt* resistance in PBW populations since 2010. *Bt* cotton is still effective on other Lepidopteran pests but the only way to manage *Bt*-resistant PBW is through re-adoption of IPM practices, more specifically cultural practices to break the pest cycle and cultivation practices like choosing short-duration cotton hybrids, early termination of cotton crop in case high-level PBW infestation, discouraging 're-flush' cropping in PBW endemic areas, mating disruption through lures and constant scouting to arrive on insecticide spray decisions. Pest management through IPM is only successful only if adopted on a wide-area scale like an entire village or a cluster of villages with contiguous cropping of *Bt* cotton. Notwithstanding the challenges in wide-area adoption of IPM it is the only hope to deal with the current PBW issue in India. Finally, it would be prudent on the part of farmers to cultivate *Bt* cotton only as a component in a compatible IPM module for an ecologically and economically sustainable cotton cultivation.

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Retaining Early Formed Squares Will Lay a Robust Foundation for Pink Bollworm Management in India

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The pink bollworm (*Pectinophora gossypiella*) (PBW) has emerged as a major menace in India in the past 10 years (Figure 1). The worm is now feeding on BG-II cotton bolls because it has developed high levels of resistance to *Bt*-cotton in India. Several strategies have been recommended for its management (Kranthi, 2015). While these strategies are important in reducing PBW damage, they can be made more effective when coupled with one main strategy of 'retaining early formed squares' This paper explains the science behind the recommendation. It describes a simple set of novel strategies to retain the first formed squares in high density planting, which not only helps to obtain high yields, but also helps the peak boll stages to escape the infestation peaks of the worm.

I would like to explain a little bit more as to why the strategy of retaining early formed squares can help the crop to escape pink bollworm damage.

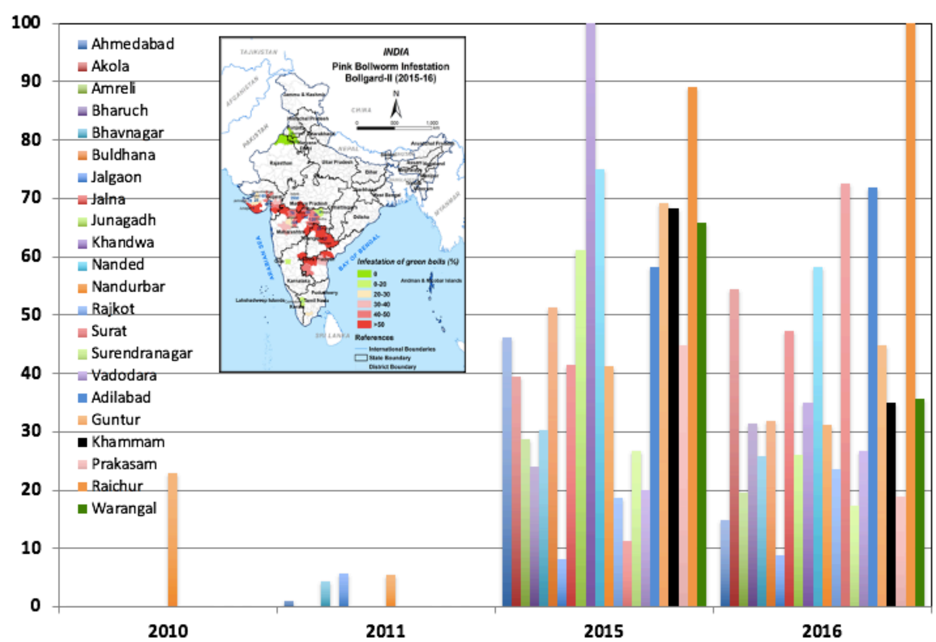

Figure 1. Recovery of PBW larvae from Bollgard-II *Bt* cotton in India

The strategy is based on the following four main scientific observations:

1. Research conducted by the ICAR-CICR and the AICRP clearly shows that the pink bollworm is a late season pest in India and late formed bolls are damaged mostly. In north India, the PBW populations appear in mid-September to assume a peak that generally occurs by mid-October; in central India PBW infests in November and peaks in early December; in south India the worms appear in December and reach peak levels by the beginning of January (Fand et al., 2021). Based on the recommended sowing months of April-May in north India; June-July in central India and July-August in south India, it can be surmised that in all the three zones, pink bollworm infestation on green bolls mainly starts after the plants attain 130 days of age. Green bolls formed within 130 days of the plant growth are generally safe from the pink bollworm. Data show that the worm occurs in 4-5 generation cycles in the cotton season starting with very low populations in the first generation that coincides with peak flowering stage, gradually leading to the third and fourth generations that cause maximum damage in late formed bolls.
2. PBW pestilence is high if the seasonal duration of preceding cotton crop was six to eight months or more. Cotton crops harvested within 150 to 160 days and terminated immediately, seldom get affected by the pink bollworm.
3. PBW neonate larvae prefer green bolls that are younger (Liu et al., 2002) that are 14 to 21 days old (Van Steenwyk et al., 1976; Henneberry and Clayton, 1982). Green bolls grow to their full size in 21 days after which they are generally considered to be relatively safer from fresh bollworm attack. It takes 45 to 50 days for a square bud to reach the stage of a full-size safe green boll. Tender green bolls are most vulnerable to fresh bollworm infestation when they are one to three weeks old.
4. In general, a 40 days old plant starts producing square buds. On average one square bud is produced per day: the production rate being dependent on genetics and environment. Presuming that all squares are retained, and no squares are shed, a plant at 130-days of age is expected to produce about 40 full-sized green bolls that are safe from fresh infestation by PBW. The plant could also have about 10-15 younger green bolls and will produce more squares/flowers and younger bolls subsequently that are most likely to get caught in PBW infestation.

Square and boll shedding are common phenomena in cotton fields across the globe. It is generally accepted that despite best management practices it may be possible to retain about 50% healthy open bolls from the squares produced by a plant, after 30% square shedding and 30% shedding or damage of the bolls that are formed out of the remaining squares. Therefore a 130-days old plant may have at best about 20 retained full-sized bolls that are safe from fresh PBW attacks and 8-10 younger green bolls that will be vulnerable to PBW infestation. Because PBW is a cryptic (hidden) pest, the crop, which is vulnerable to PBW after 130 days, can only be partly protected from PBW damage by resorting to integrated pest management strategies that include pheromone mass trapping using the traps and prolonged-effect lures developed by ICAR-CICR. The lures can also be used for mating confusion. Whenever necessary, insecticides such as ememectin benzoate, flubendiamide, chlorantraniliprole or spinosad or indoxacarb or novaluron or thiodicarb (http://www.aiccip.cicr.org.in; Divya *et. al*., 2020) can be used at economic threshold levels of 8 moths per trap per night for three consecutive nights.

Figure 2. Pink bollworm larval damage

Figure 3. Pink bollworm damaged rosette flowers

Figure 4. Pink bollworm pupa

Figure 5. Pink bollworm moth

Table 1. Predicted Crop Phenological Stages in Central India

Date	Age of the crop	Phenological stage
15-Jun		Sowing
20-Jun	0	Seedling emergence
30-Jul	40 days old	Square formation starts
10-Sep	80 days old	40 squares are produced per plant
05-Oct	105 days old	40th square blooms into a flower
30-Oct	130 days old	40th square produced by the plant will have turned into a full-size green boll that is safe from a fresh bollworm attack. The chances are that from the 40 squares produced, only 20 green bolls or less will be retained per plant.
01-Nov	131 days old	Pink bollworm starts attacking tender green bolls (<20 days old). Green bolls produced after mid-October are tender and vulnerable to a fresh attack by the pink bollworm in November and later.
01-Dec	161 days old	PBW populations reach a peak and cause significant damage

Data shows that in central India, pink bollworm infestation starts in November and reaches a peak by late November to early December. Green bolls that reach full-size by the end of October escape the pink bollworm and green bolls that are formed later, which are less than 20 days old are vulnerable to PBW oviposition and infestation. Green bolls formed before the end of October are also likely to receive the benefit of residual soil moisture and available nutrients depending on moisture retention capacity of the soil and the seasonal rainfall pattern. Under a normal monsoon pattern, most soils in rainfed farms become dry by the end of October and late formed bolls suffer stress. Bolls harvested from the shorter season crop are healthier and produce clean good quality fibres, because they are rarely starved of water and nutrients and also because they escape PBW infestation.

Cotton plants compensate for square shedding. Shedding of early formed squares prompts plants to shift towards vegetative growth by producing new fruiting branches and new squares in efforts to compensate for the lost squares and bolls. However, compensation needs energy; it leads to elevated requirements of water and nutrients thereby accelerating stress; further loss of fruiting parts and a longer crop duration. A longer season leads to late formed bolls that are most vulnerable to PBW infestation, which in turn leads to a higher number of PBW generations and a need to extend the crop to recover lost yield. A longer season thus supports higher pestilence in the current and the subsequent cotton crop.

Research clearly confirms that 'retaining early formed squares' enables higher 'water-use-efficiency', better 'nutrient-use-efficiency' and efficient energy partitioning without subjecting plants to any additional stress. Further, higher yields can be obtained from a timely sown, shorter season crop of 150 to 160 days by combining the strategy of 'retaining early formed squares' with high density planting (see explanation below).

Technologies that help to retain early formed squares and bolls may optimally enable a retention of 50% healthy bolls from the total number of squares produced and at worst enable 30% retention. Data indicates that low yields in India and Africa are mainly due to low density of plant population coupled with higher rate of square/boll shedding/damage which could reach as high as 80 per cent or even more.

How can high yields be obtained from a short season crop of 150 to 160 days?

High yields can be obtained from any crop irrespective of the level of plant population density, by preventing shedding of early formed squares and bolls to the best extent possible. However, to obtain the same level of yield, a crop with low density plant population will require a longer duration, while a crop with a higher density plant population will require a shorter duration depending on the density levels. I am presenting two tables below to explain how plant population density influences the crop duration to harvest the same level of yield and how high yields can be obtained from a short season crop of 150 to 160 days.

Table 2. Yields (Kg/ha) from a short season crop of 150 to 160 days at different plant densities

Plant spacing (90 cm between rows)	Plant population per hectare	Number of healthy bolls retained per plant on 130 days old plants, that escape PBW infestation	*Calculated lint yield Kg/ha @ 20 healthy bolls retained per plant on 150 to 160 days old crop
90 x 90cm	12,345	20	247
90 x 60cm	18,518	20	370
90 x 45cm	24,691	20	494
90 x 30cm	37,037	20	741
90 x 15cm	74,074	20	1481
90 x 10cm	111,111	20	2222

*The average lint weight in Indian bolls is about 1.3g. For calculation purposes of a worst-case scenario, each open boll was assigned a value of 1.0g lint.

Table 2 shows a hypothetical case as an example where technologies are used to ensure retention of 20 healthy full-formed bolls from 40 squares formed on a 130-days old plant by retaining 70% of the first formed squares and 70% bolls that resulted from the retained squares. At a low plant density of 12,345 plants per hectare, with each boll providing 1g lint, the calculated yield is 247 Kg lint per hectare from a 150-160 days old crop. In stark contrast, the calculated yield would be 2,222 Kg lint per hectare from the same duration of 150-160 days old crop at a plant population density of 111,111 plants per hectare.

Table 3. Number of retained healthy bolls required to get 778 kg lint/ha (2018 world average) and calculated crop duration for the target yield at different plant densities

Plant spacing (90 cm between rows)	Plant population per hectare	Number of healthy bolls (@ 50% retention), required to get 778 kg lint/ha	Calculated crop duration (days) to get 778 kg lint/ha at 50% boll retention
90 x 90cm	12,345	62	247
90 x 60cm	18,518	42	205
90 x 45cm	24,691	31	184
90 x 30cm	37,037	21	162
90 x 15cm	74,074	10	141
90 x 10cm	111,111	7	134

Table 3 estimates the number of bolls and the duration of the crop required to achieve a target of the world average lint yield of 778 Kg/ha., at different plant densities. The table highlights the need for a longer duration of 247 days and a higher number of 62 healthy bolls at 50% retention rate at a low plant population density of 12,345 plants per hectare to harvest a target lint yield of 778 Kg/ha, in stark contrast to high density planting system of 111,111 plants per hectare which needs only 7 bolls per plant and just 134 days to achieve the same target yield of 778 Kg/ha. Thus, fields with low density of plant populations require retention of a greater number of squares per plant to obtain the same target yield. Retention of a greater number of squares and bolls per plant means a longer seasonal window for similar yields that can also be obtained from a shorter season with higher density of plants per hectare.

A long duration crop is not desirable from a management perspective because it mandates a long vulnerable management window. A longer window of flowering and tender green bolls imposes higher challenges for a longer vigilance-window to provide adequate water, nutrients and protect the crop against bollworms. For example, to retain 127 squares for 62 bolls per plant at 49-50% retention, it would require a vigilant management window of up to 177 days to minimize shedding, starting from the square initiation stage until ensuring the safety of the last batch of bolls. On the other hand, retaining 14 squares for 7 bolls per plant would require an initial vigilant management window of only 64 days to minimize shedding starting from the square initiation stage until ensuring the safety of the last batch of bolls. A longer season is a recipe for disaster in terms of crop management and bollworm management, especially because it necessitates higher use of water, fertilizers and pesticides apart from creating a perennial cyclic problem of the pink bollworm.

Retaining early formed squares and bolls

Squares are formed sequentially on the fruiting branch, first at the first position node (see figure 1 below), followed by the next square on the second position approximately after six days (depending on genetics and environment) and so on. (Figure 6)

The first position square/bolls are most favoured by the plant for nutrition and water, followed by the second and third position fruiting parts. Data show that the first and second position bolls have the best quality fibre because they receive a preferential treatment. Shedding of these fruiting parts imposes high levels of stress to the plant. Research across the world showed that bolls at the first, second and third position of the fruiting branches contribute most towards harvestable yields at about 60%, 30% and 10% respectively. Therefore, retention of at least the first and second position squares/bolls, which represent the early formed fruiting parts, is crucial for high yields.

Figure 6. Position of fruiting parts on a fruiting branch

Minimizing shedding of early formed squares/bolls results in higher production efficiency of plants, synchronous early maturity, escape of damage by pink bollworms, high yields in a short season and facilitates timely termination of the crop.

Formation of fruiting parts depends on ideal conditions of heat, light, water, nutrients and absence of biotic and abiotic stress. Early formed squares are shed mainly due to one or more of three major factors:

- Canopy-shading or cloudy conditions or waterlogging or drought or extreme temperatures
- Deficiency of nitrogen or phosphorus or boron
- Insects such as plant bugs, mirid bugs or bollworms
- Square shedding can be effectively minimized by using any of the following technologies

Chemical sprays to interfere with abscission

A number of chemicals that interfere with abscisic acid and ethylene levels in the plant have been used as foliar sprays early in the season to minimise physiological square shedding. For example, spray of 1-Naphthalene Acetic Acid (NAA) @ 40ppm during early square formation stage has been found to minimize physiological square shedding. Ethylene inhibitors such as Aminoethoxy Vinyl Glycine (AVG) and 1-Methyl Cycloprene (MCP) have also been found to minimize physiological shedding of squares and bolls (Brito et al., 2013; Najeeb et al., 2015). Several other chemicals such as Amino-oxy-acetic acid (AOA), Triacontanol, 2,3,5-Tri-Iodo-Benzoic Acid (TIBA), Silver thiosulphate, Silver nitrate and Trans-cyclo-octene have been tested across the world in cotton for their role in inhibiting square and boll shedding (Patel, 1993; Freytag and Coleman, 1973; Prakash et al., 2007; Tariq et al., 2017). There is a need to validate their dose and application at proper growth stage under local conditions.

Canopy management

Mepiquat Chloride (15 to 30g a.i/ha) is commonly used in developed countries at 50-80 days after sowing at bi-weekly interval (Cook and Kennedy, 2000; Srivastava, 2002) for canopy management at thresholds of >4.0cm average internodal length of the main stem to prevent canopy-shading. Canopy management in the early stages of square formation is crucial for proper light penetration to reduce shedding of squares and early formed bolls. Alternatively, Paclobutrazol (40g a.i/ha) can also be used as one or two applications during 50 to 80 days for canopy management and to prevent square and boll shedding (Temiz et al., 2009; Choudhary and Choudhary, 2016).

Nutrient management

Application (basal dose or foliar sprays at early squaring stage) of nitrogen / phosphorus / boron based on soil fertility helps in minimizing square and boll shedding.

Boron application

Boron plays an important role in square and boll retention. A list of cotton growing districts where majority of farms were reported to be boron deficient, is presented below (Table 4). Boron must be applied in fields where it is reported to be deficient. Depending on the deficiency, Borax must be applied as band placement at 10 to 20 Kg/ha at the time of planting and if necessary, as foliar sprays of 0.1 to 0.3% on 40-80 days old crop to minimize square shedding.

Soil Moisture management

Draining of waterlogged fields and providing irrigation as and when required by the plants helps in minimizing square shedding.

Insect pest management

Plant bugs, mirid bugs and bollworms cause square shedding. Bugs can be controlled using selective insecticides such as

Table 4. Districts where boron deficiency has been recorded in majority of the farms tested

Maharashtra	Jalna, Nagpur, Nanded and Satara
Punjab	Bhatinda and Patiala
Karnataka	Bagalkot, Belgaum, Bellary, Bidar, Bijapur, Chikballapur, Chikmagalur, Gulbarga, Hassan, Haveri, Koppal, Mysore, Tumkur and Uttar kannada.
Madhya Pradesh	Betul, Dhar and Neemuch
Tamilnadu	Coimbatore, Cuddalore, Dindigul, Erode, Madurai, Namakkal, Sivaganga, Tiruchirappalli, Tirunelveli, Tiruvannamalai, Tuticorin, Villupuram and Virudhunagar
Telangana	Nagarkurnool and Rangareddy
Haryana	Sirsa
Odisha	Anugul, Balangir, Boudh, Ganjam, Kalahandi, Kandhamal, Kendujhar, Koraput, Nabarangpur, Nayagarh, Nuapada and Sonepur
Gujarat & AP	No data

Azadirachtin-based insecticides or Diafenthiuron or Buprofezin or Flonicamid. Early season bollworm infestation can be efficiently controlled with biological control or Indoxacarb or Chlorantraniliprole or Spinosad or Flubendiamide or Emamectin benzoate at doses and ETLs recommended by ICAR-CICR (http://www.aiccip.cicr.org.in). These insecticides are relatively selective with higher toxicity to target pests and lesser toxicity to beneficial insects. (Table 4)

Other strategies of PBW management and high yields from a short season

Basically, all cotton varieties across the world are indeterminate which means genetically ordained to produce bolls continuously for multiple picking. A combination of genetics, agronomy and physiological techniques is used to orient the crop towards single picking. It is with these technologies that countries such as Australia, China, Brazil, Turkey, Mexico and USA are able to harvest high yields of 1000 to 2000 Kg lint per hectare in a short season of 5-6 months compared to the average Indian yield of 500 Kg/ha in 7-8 months. High yields of 1000 to 2000 Kg/ha are obtained in Brazil, Australia, Mexico, Greece, Spain and USA through single picking and in China through multiple picking. These countries use high density planting coupled with canopy management and protection of early formed squares so that the crop duration is kept short (5-6 months) and high yields can be harvested within a single synchronous picking. Brazil is almost completely rainfed and USA cotton is 35% irrigated just like India. Indian weather is actually best suited for cotton compared to these countries. However, Indian average yields are only as good as a few resource-poor rainfed Africas countries such as Cameroon, Cote D'ivoire, Benin and Mail, which nether have *Bt* nor Hybrid cotton and use less than one-fifth of the fertilizers that India uses.

Following is a list of five main strategies that are critical for India to harvest high yields and move towards a robust long term PBW management system.

1. **Area-wide cotton-free closed season of 6 months**

   While it is important that India explores options of short season cotton (5-6 months) and enables an area-wide cotton-free 'closed season' of 6 months, four other strategies also play a critical role in PBW management. Pink bollworm feeds mainly on cotton and a six months cotton-free period hits it very hard. Timely termination of the crop within 5-6 months helps the crop to escape PBW which is primarily a late season pest that starts its main infestation after mid-November. A closed season significantly reduces pest carry over to the next season. Though timely termination and closed season play an important role in minimizing pest carry over of normal PBW populations that will not survive in the absence of food, the presence of diapausing larvae that are dormant during the cotton-crop-free period, presents a serious challenge.

2. **Destruction of unharvested boll and gin waste**

   Some of the PBW larvae enter diapause at end of the season. These larvae pupate and moths emerge in the subsequent season when the crop starts flowering. Most of the diapausing larvae take shelter in unharvested immature bolls and also inside seeds. Very few diapausing larvae drop on the soil. Therefore, the second most important strategy is to destroy all residual unharvested bolls and also destroy gin waste which contains diapausing larvae.

3. **Retention of early formed squares**

   Retain early formed squares (flower-buds) through canopy management, controlling mirid bugs, application of Boron sprays wherever deficiency is prevalent and naphthalene acetic acid (NAA) sprays to combat physiological stress. More than 60-70% of early formed squares are shed in India and the crop stretches itself into a long season in its attempts to compensate the early losses.

4. **Early season mass trapping and mating confusion**

   Use mass trapping and mating confusion techniques during early flowering stage so that the first generation PBW moths are least productive. A new slow-release pheromone formulation was developed (Kranthi, unpublished) and commercialised as a mass trap that attracts and traps male moths for 50 to 60 days without the need to change lures. The pheromone lures and traps can be procured from Innovative Biosciences Nagpur or Central Institute for Cotton Research Nagpur. Studies showed that 50 traps per hectare were effective is mass trapping moths in the early stages of the crop to reduce subsequent pestilence. Mating confusion technique requires at least 500 PBW pheromone ropes or dispense spots. For small holder farming conditions, mass trapping could be a better option compared to mating confusion not only because it is much inexpensive, but also because it is possible that the moths from pheromone treated plots are likely to disperse into neighbouring pheromone-untreated fields.

5. **Pheromone monitoring and control**

   Pheromone traps offer an elegant option for PBW monitoring. Control measures can be initiated at an economic threshold levels (ETLs) of eight moths per trap per night for three consequent nights, with eco-friendly insecticides, biopesticides and biological control. The following interventions, namely application of azadirachtin or *Trichogramma bactrae* or chlorantraniliprole, spinosad, flubendiamide, emamectin benzoate, indoxacarb etc., were found to be effective and relatively less toxic to naturally occurring biological control compared to conventional insecticides belonging to the classes of organophosphates and synthetic pyrethroids.

These five strategies have the potential to lay a firm foundation for PBW control.

The feasibility of adopting ‘mating confusion’, ‘sterile insect release techniques (SIT)’ ‘refuge-in-bag’ ‘new *Bt* genes’ and ‘short season’ strategies for PBW management in India and Pakistan

An intensive discussion has been taking place in India and Pakistan to explore the best options for long term management of PBW. At least strategies, namely ‘mating confusion’, ‘sterile insect release techniques’ ‘refuge-in-bag’ ‘short season’ and novel genes based *Bt*-cotton are discussed most frequently.

Mating confusion and SIT

Sterile insect release technique and pheromone-based mating confusion techniques together are believed to have contributed immensely in PBW eradication in USA and Mexico. Both these techniques require area-wide implementation and are very expensive. The release of billions of male sterile moths has to be essentially driven by a Government policy that is approved by farmers. The mass production of sterile moths and regular aerial releases are extremely expensive and appear to be improbability in India and Pakistan. Pheromone based mating confusion requires at least two releases of 500 PBW pheromone ropes per hectare or 2-3 applications of Specialized Pheromone & Lure Application Technology (SPLAT) are expensive and appear to beyond the reach of small holder farmers. Further, there are very few studies that better our understanding as to whether the confused male moths remain in the pheromone-treated field or disperse to the neighbouring pheromone-untreated fields in search of females to create more pestilence.

Refuge-in-bag

A non-*Bt* cotton refuge has the potential to delay resistance when it has not surfaced or when it is at a low level. However, refuges will not be able to reverse PBW resistance to *Bt*-cotton or minimize pest damage. The Government of India in an official gazette notification (December 2016) stipulated that the *Bt* seeds in a ‘refuge-in-bag’ should be between 90 to 95% and the isogenic non-*Bt* refuge cotton seeds of the corresponding *Bt*-hybrids must be between 5 to 10%. Under the given conditions, the ‘refuge-in-bag’ strategy will not make any tangible difference to the prevalent ‘PBW resistance to *Bt*-cotton’ nor will it be able to strengthen PBW management in any manner.

New Bt-gene transgenic cotton

Tabashnik (2021) suggests the use of *Bt* genes Cry1B and Cry1C for new transgenic cotton. It remains to be seen if PBW which is a functionally monophagous pest will remain susceptible for long to the new transgenes, Cry1B and Cry1B if the ecological conditions of long season cotton continue to be the same as they are today.

Area-wide short season and closed season

A short season is critical for an area-wide closed season. Indian farmers and seed companies generally believe that high yields are possible only with a long season. Harvesting high yields with short season is in many countries has been possible only with high density planting (90x10cm) provided the early formed squares are retained at least to 70-80%. Hybrid cotton is not very conducive for high density planting because of the costly seeds. Therefore pure-line varieties are a better option as is the case in the countries that harvest more than 1000 Kg lint per hectare. Turkey, Greece and Spain grow non-*Bt* varieties and harvest 1000 to 1700 Kg lint per hectare with 3-6 applications of insecticides. There is no reason to believe that this will not be possible in India and Pakistan.

The short season high density cotton presents advantages and disadvantages. There are at least three advantages. The HDPS single pick system enables to obtain high yields within a short time, because the green boll formation window which is most critical for pest and nutrient management is short (40-50 days), compared to the longer window (40-120 days) as is the case now in India. Fibre quality of early picked synchronous bolls is relatively uniform and much better than the late picked bolls. Short season cotton if grown in an area-wide manner to ensure an area-wide closed season of 5-6 months will certainly bring down PBW populations significantly. The disadvantages could be mainly due to drought and the difficulty of criss-cross hoeing for weed management, which is the main method of weed control in India. Drought poses the biggest threat to short season systems which in some cases may not provide congenial conditions for the crop to compensate a severe damage. Management in cropping systems designed for a short season becomes critical if the crop gets damaged in the early stages due to drought effects, flooding or insect infestation. Weed management between plants will need extra attention because criss-cross hoeing is not possible in high density systems because of the narrow 8-10 cm spacing between plants. Studies (Kranthi, unpublished) showed that weeds can be managed with hoeing between rows and if necessary, with application of any selective herbicide between plants.

Conclusion

The pink bollworm is a monophagous pest with cotton as the primary host. A ‘closed season’, where no cotton or PBW alternate host crops are allowed to be grown between two cotton seasons, is almost universally enforced, wherever cotton is cultivated to prevent carry over of PBW from the previous crop. Currently strict adherence to this ‘closed season’ is one of the most effective methods available for the control of pink bollworm in Africa and a key IPM strategy across the world. Deployment of ‘short season’ and ‘closed season’ are the most common universally recommended strategies for PBW management. These two strategies along with high density planting were used to produce high yields and effectively combat the serious menace of PBW in the desert valleys of southern California and Arizona in the mid-1970s to the mid-1980s; and would be most applicable to combat the current PBW crisis in India. Pink bollworm is known to cause least problems in countries that cultivate short-season cultivars and implement a closed-season of at least 5 to 6 months. Therefore, the best strategies for PBW management in India would be to ensure timely sowing in an area-wide manner and retention of early formed squares in high density planting so that high yields can be obtained from a short season crop; destroy unharvested bolls and seeds harbouring diapausing larvae; monitor and manage the first 1-2 generations of PBW through mass trapping and eco-friendly insecticides and timely termination of the crop.

Photograph: Mahesh Upender

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