BIOCONTROL BASED COTTON IPM

S.Vennila

Biocontrol is a constitutive component of IPM that provides sound ecological foundation for sustainable cotton production. Also referred as biological control, the pest suppression is achieved by the action of living organisms on pests. The principle behind biological pest control is that a given pest has enemies – predators, parasites or pathogens. By introducing or encouraging such enemies, the population of pest organisms decline. Biocontrol in cropping systems operate in three ways *viz.,* Natural, conservation and augmentative biological control.

Natural biological control:

Beneficial insects growing wildly in fields are one of the natural sources of pest management. When beneficials are abundant in cotton field, higher pest levels can be tolerated for longer periods without pesticide use, which means there is savings of pest control cost. A number of naturally occurring native predators such as coccinellids, chrysopids and syrphids besides many parasitic wasps offer significant control of early season sucking pests such as jassids, aphids and thrips.Hymenopterous and tachinid parasitoids (e.g. *Eriborus argenteopilosus, Compoletis chlorideae, Microchelonus* spp. *Palexorista laxa, Carcelio illota* and *Goniopthalmus halli*) are common on *Helicoverpa armigera. Rogas aligarhensis* parasitisation on *Earias vittella* and pink bollworm control by *Apanteles angaleti* and *Bracon greeni* are quite frequent.Natural mortality of semilooper and *H. armigera* due to fungal pathogen *Nomuraea rileyi* during cooler months and years of epizootics is an observed phenomenon. Many species of spiders and birds also execute a fair amount of natural control of cotton insect pests. All these native natural enemies are adapted to local environment and to target pest, and hence are very effective. List of important native natural enemy groups recorded on various insect pests along with their host stage attacked are given in Annexure 1.

Conservation biological control:

Second approach of conservation biological control involves identification and modification of factors that limit the effectiveness of the natural enemies. Slight modifications in farming practices and ecological planting schemes can create cotton agro ecosystems more closely emulating natural ecosystems and promote in field populations of voracious pest eating beneficials at no additional cost to farmers. Cotton field populations of dominant natural enemies are markedly high when cotton agro ecosystem diversity is increased as compared to monocultures. Intercropping with cowpea was found to increase coccinellids, and parasitism of spotted bollworm under south Indian conditions. *H. armigera* parasitism by hymenopterous parasitoids in central India was high when late variety red gram was grown as stirp or border crop with cotton. Interplant maize and cowpea act as a source of predators against *H.armigera*. Factors that contribute to high level of natural enemies in diversified ecosystem are the availability of diverse microhabitats, greater availability of food sources (prey, nectar,pollen), alternative hosts and shelter all of which encourage colonization and population build up of natural enemies. The refuge or source function of border maize or cowpea are attributed to the abundance of floral nectar and alternative prey (aphids), shelter, mating and oviposition sites harbored in the border crop compared with monoculture cotton having lesser biodiversity. In some situations, this may include reducing the application of pesticides, since such pesticides may kill predators at the same time as killing the pests. Sometimes part of a crop area is left untreated so that natural enemies will survive and recolonise the treated areas. Other non-insecticidal methods of pest suppression are given precedence over use of insecticides towards conservation of natural enemies. Erecting bird perches increases the visitation by the birds and hence their predation on insects.

Augmentative biological control:

Augmentation is be achieved by mass production and periodic release of natural enemies of the pest, and by genetic enhancement of the enemies to increase their effectiveness of control. Natural enemies that are amenable to mass-production for use in cotton ecosystem are: the egg parasitoid,

Trichogramma, a generalist predator, *Chrysoperla*, pathogens such as *Helicoverpa* Nuclear Polyhedrosis Virus (H-NPV). Heat tolerant and multiple insecticide tolerant strains of *Trichogramma* for use in cotton ecosystem are available. Other pathogens such as *Verticillium lecanii* against aphids, *Metarhizium anisopleae* against jassids, *Beauveria bassiana* against whitefly, *Nomuraea rileyi*, *Entomophthora* sp.and entomopathogenic nematodes *Heterorhabditus* and *Steinernerma* spp. against bollworms have considerable promise in cotton IPM.

Mass production and field use of bio agent *Trichogramma chilonis*

Trichogramma chilonis is an egg parasitoid and agent under biological control of lepidopteran insect pests. It is effective against bollworms of cotton. Since it attacks the egg stage, damage done by larvae is avoided. Mass production of the parasitoid requires laboratory culture of a host insect and a nucleus culture of the parasitoid.

Steps involved in production rearing of host insect

Equipments/materials required for mass production of *C. cephalonica*

Non recurring items	*Recurring items*
• Hot air oven • Slotted angle iron Racks • Corcyra rearing boxes (Plastic basins) • Refrigerator • Oviposition cages (modified plastic fennels) • Vacuum cleaner • Grinder & Blenders • UV light source	• Crushed Sorghum grains • Yeast • Honey • Formalin • Proteinex • Vitamin E • Ground nut • Measuring cylinders • Seives/ brushes • Cotton cloth

Corcyra cephalonica has been used for mass multiplication of *T. chilonis*. The nucleus cultures can be eggs of *Corcyra* or larvae, which can be obtained from research institutes maintaining cultures. Sorghum grains are used as feed material for the larvae. Sorghum grains should not be treated with insecticides.The requisite quantum of sorghum is milled to get 3-4 pieces of each grain. Sorghum grains are heat sterilized in oven at 100ºC for 30 minutes and the grains are sprayed with 0.1% formalin. This treatment helps in preventing the growth of moulds as well as to gain humidity, which was lost due to heat sterlisation. Then grains are air dried. In each rearing container, 2 kg of sorghum grains are filled and inoculated with 0.125 cc, i.e, (2500 eggs) of mother culture. Date of inoculation should be written on larval rearing containers, to know the duration of each container is being used. Bakers yeast @ 5 gm per container is added to enhance egg laying of the adult moths and ground nut @ 10 gm/basin is also added for enriching the diet. After about 40 days of inoculation, moths start emerging and the emergence continues for two months. 10 to 75 moths emerge daily with the peak emergence being between 65th and 75th day. Moth emergence reduces after 100 days of initial inoculation and basins are emptied thereafter. Moths are collected daily (by using mechanical moth collection device like vacuum cleaner) and transferred to plastic buckets containing cotton swabs saturated with honey +vitamin E +Proteinex mixture and covered with cotton cloth for mating and oviposition. Roughly 2000-3000 pairs of moth can be placed in one chamber. Eggs from the buckets are collected, sieved and are used for inoculation in to the feed material for the larval and pupal stages to develop from which the moths emerge or diverted for production of trichocards. Eggs collected manually are placed in tubes and quantified with a measuring cylinder. Approximately 1cc contains about 16,000 to 22,000 *corcyra* eggs at the fresh harvest.

Production of Trichocards

The eggs of *Corcyra* thus collected are cleaned to make it free from insect scales. They are sieved thrice and then poured on a plain paper. By tapping on one side of the paper the round eggs comes downward and flat surfaces stick to on the paper. Thus, the cleaned eggs are spread on the gummed cards (9 cm x 22 cm) with the help of screen. These eggs of *Corcyra* are exposed to UV radiation

with 15 watt UV tube for 45 minutes for killing the embryo of the eggs. The eggs exposed distance such that the egg card is about 12 to 15 cm from the UV source. However, the eggs meant for further multiplication of *Corcyra* should not be treated with UV rays. After the sterlisation, the egg cards are placed in polythene bags, and are exposed to nucleus culture of Trichogramma adults. The ratio of *Trichogramma* adult to host eggs should be maintained at around 1: 6. Parasitisation of *Corcyra* eggs take place in 24 hours of exposure to the mother culture of *Trichogramma* sp. The egg turns black after three days from the day of parasitisation. The trichocards, the cards on which parasitised *Corcyra* eggs are glued, should reach the field within 3 days. The parasitised eggs can be kept in refrigerator at 10ºC for about 12 days. The percentage of parasitisation received over host eggs ranges from 62% to 90%.

Field release of Trichocards

The trichocards are released in the field 45 days after sowing @ 5-8cards/ha/release (one to one and a half lakh eggs). Totally two to three releases are made depending upon the build up of pest population coinciding with the egg stage of the target pest.
The following precautions are required to be taken while using trichocards:

- Trichocards should be packed in such a way that the parasitised surface is on the inner side.
- Emergence date should be specified on cards for the guidance of the users.
- Cut pieces of 'trichocards' should be stapled on the inner-side of the leaf to avoid direct sunlight.
- Cut pieces of the card should be stapled in morning hours and just before emergence to avoid predation.
- Farmers should refrain from using pesticides in the field where *Trichogramma* are released. If need arises selective/safer pesticides can be used and it is to be ensured that pesticides are used 15 days before or after release of *Trichogramma*.

Production of Chrysoperla

In mass production of *Chrysoperla* sp, the larvae are either reared in plastic tubes or empty injection vials or in groups in containers or in individual cells. In the present model the larval rearing is done in plastic injection vials, which can be procured easily at a cheap rate. Field collected larvae of chrysopa or mother culture of chrysopa are kept in injection vials and corcyra eggs, which act as a food material for *Chrysoperla* sp, are placed in vials,. Recent research reveals that synthetic diet consisting of soyabeans can replace *Corcyra* eggs for efficient and cheap production of *Chrysoperla* eggs. It is assumed that 1cc of *Corcyra* eggs are required for rearing 100 larvae and final yield of adults are assumed at 50, considering the predation and mortality factors. It takes 15 days to pupate and matured larvae spins a cocoon from which adult emerges in 5-7 days. Adults on emergence mate repeatedly. The mature adults are transferred to the oviposition cages, which could be modified plastic dust bins containing open surfaces at both ends covered with nylon nets. The adults are fed daily using, cotton swabs, which contains drinking water, Honey, protinex, fructose and castor pollen. In each oviposition container roughly 20 pairs can be accommodated and inside portion of the container is covered with black paper on which adults lay eggs. The productive adult life has been conservatively assumed at 200 though it can go upto 600-800 eggs per female, as it is determined by the management practices, particularly food and climatic factors. Black paper containing eggs can be cut into bits, for door to door supply to the farmers.One to two thousand eggs per hectare are recommended for field release coinciding with jassid,aphid and/or bollworm occurrence. Small bits of egg card containing at least 10 eggs are stapled on to the under surface of the top canopy of the crop.

Mass production of *Helicoverpa* nuclear polyhedrosis (HNPV) virus:

Mass production of HNPV can be done in two ways: 1) through the use of field collected larvae and 2) under laboratory. For mass production through field collected larvae second instar larvae of *Helicoverpa* are collected from the field. Larvae are reared on artificial diet prepared and kept in multicell plastic trays. To prepare artificial diet 105g of Besan, 2g of methyl para hydroxy benzoate, 1g of sorbic acid and 10g of yeast are added and blended along with 390 ml of water in a mixer for two minutes. 10 g agar-agar is boiled with another 390 ml of water and added to the mixer and again

blended for one minute. 3g of ascorbic acid, two capsules each of vitamin E and multivitamin complex, 2ml of 10% formaldehyde solution and 0.25g of streptomycin sulphate are added at last and mixed for half a minute. Diet of semi solid consistency is immediately poured into the multi cell trays and allowed to cool and solidify. Inoculums of HNPV are added to the diet and larvae are allowed to feed for 4-5 days. The infected larvae die after a week and they are collected in water and mixed well. Centrifugation is done to remove the debris and to retain only the virus particles.
For HNPV production under laboratory , field collected larvae are allowed to grow and pupate. The pupae are sterilized in 0.1% sodium hypo chlorite solution and set aside in plastic jars for moth emergence. On moth emergence mal –female moths in 1:1 ratio are released into oviposition cages having hanging muslin cloths. Moths start laying eggs after 3-4 days and continue to lay eggs for a week. Muslin cloth is replaced once in two days and cloth with eggs are incubated for worms to emerge. The larvae are transferred to diet prepared as given above for infecting them. 20% of the larval population should be reared on diet without contamination with virus to maintain host culture continuously. For getting purified virus the cadavers are macerated in mixer followed by centrifugation at 5000 rpm for 30 minutes. The supernatant contain the virus and are filled after calibration of polyhedral inclusion bodies(PIB) per ml. Normally 250 larval equivalents(LE) (1LE=2X10^9 PIBs) per hectare are used for control of *H.armigera*. Application should be targeted against the early instars of the pest for its efficacy. Application of HNPV is to be advocated at evening hours.

General points for consideration:

- Bio agents are largely specific to pests and their stages.
- Timely availability and application is mandatory to harness their efficacy.
- A supplemental release at times of activity of native natural enemies proves highly effective.
- Reduced and selective pesticide use aids in sustaining the released bioagents.
- Use of bio agents fits well in to the organic cotton production systems.
- Too many releases prove to be cost ineffective.
- Application at times of heavy rains cause loss of inoculum in case of parasitoids.

Quality issues

In any rearing programme, it is important to maintain and improve the quality of the bio-agents produced. It is important to maintain the viability of the culture, start with a genetically diverse population, avoid deleterious selection in the lab and encourage desirable genetic changes. However, prime importance should be given to testing the value of selected populations in the field.
For assessment of the quality of any bio-agent, two types of tests can be conducted: a) production and process tests which include factors like fecundity, per cent hatch, yield of pupae or adults, size of the progeny, etc. and b) performance tests which include searching ability, release and re-capture in field, mating compatibility, flight capacity, genetic diversity, ability to control the pest, etc.
Step to be followed in a rearing system for quality assessment and control,

a) Internal standards to be established after a period of laboratory maintenance
b) Periodic comparison to wild strain
c) Quality traits to be determined by approximate assessment of growth, survival and reproduction
d) Wild standard established based on performance of a particular stage
e) To formulate specifications for mass reared insects based on characteristics required for success in the field
f) Standards to be developed based on traits required for a potential natural enemy

Annexure 1: Native natural enemies of cotton pests

Natural enemy	Pest	Stage of pest
A. Parasitoids		
Aphelinus sp	Spotted bollworm	Egg
Erythmelus empoascae	Spotted bollworm	Egg
Gonatocerus sp	Spotted bollworm	Egg
Trichogramma achaeae	Pink bollworm	Egg
	Spotted bollworm	
T. brasiliensis	Spotted bollworm	Egg
T. chilonis	Spotted bollworm	Egg
	American bollworm	
T. chilotraeae	Pink bollworm	Egg
	Spotted bollworm	
Telenomus remus	Spotted bollworm	Egg
Trichogrammatoidae sp near *guamensis*	Pink bollworm	Egg
	Spotted bollworm	
Agathis fabiae	Pink bollworm	Larva
	Spotted bollworm	
Apanteles angaleti	Pink bollworm	Larva
Bracon chinensis	Pink bollworm	Larva
Bracon greeni	Pink bollworm	Larva
	Spotted bollworm	
Bracon kirkpatricki	Spotted bollworm	Larva
Bracon brevicornis	Spotted bollworm	Larva
Bracon habator	Spotted bollworm	Larva
Camptolithlipsis gossypiella	Pink bollworm	Larva
Rogas aligarhensis	Pink bollworm	Larva
	Spotted bollworm	
Goniozus sp	Pink bollworm	Larva
Campoletis chloridae	American bollworm	Larva
Elasmus johnstoni	Pink bollworm	Larva
Pyemotes ventricosus (mite)	Pink bollworm	Larva
Chelonus sp	Bollworms	Egg-Larva
C. blackburni	Pink bollworm	Egg-Larva
Xanthopimpla punctata	Cotton leaf roller	Pupa
Brachymeria sp.n. euploeae	Cotton leaf roller	Pupa
B. apantelesi	Spotted bollworm	Pupa
B. nephantidis	Spotted bollworm	Pupa
Encarsia formosa	Whitefly	Nymph
Encarsia shafeei	Whitefly	Nymph
Eretmocerus mundus	Whitefly	Nymph
B. Predators		
Chrysoperla carnea	Sucking pests & bollworms	Egg, nymph, adult
Brumus saturalis	Sucking pests & bollworm	Egg, nymph
Coccinella septumpunctata	Sucking pests & bollworm	Egg, nymph
Menochilus sexmaculatus	Sucking pests & bollworm	Egg, nymph
Geocoris sp	Sucking pests	Nymph, Adult
Zelus sp	Sucking pests	Nymph, Adult
Spiders	Sucking pests & bollworms	Nymph/Larva, Adult
Canthecona furscellata	bollworms	Larva
Encarsia sp	Whitefly	Nymph, Adults
Syrphus confracter	Aphids	Nymph, Adults
S. baleatus	Aphids	Nymph, Adults
S. searius	Aphids	Nymph, Adults
C. Pathogens		
Aspergillus sp.	Whitefly	Nymph
Bacillus thuringiensis	Bollworms	Larva
Beauvera bassiana	Bollworms	Larva
NPV	*Helicoverpa* &*Spodoptera*	Larva
Nematodes	Bollworms	Larva

Source: Dhawan, 1990,1999,2000,2005; Natrajan and Seshadri, 1989; Natrajan and Sundaramurthy, 1990; Rao *et al*., 1994a,; Sundarmurthy and Chitra, 1992; Puri *et al*., 1999.