ROLE OF BIOCONTROL AGENTS FOR THE PEST MANAGEMENT IN MULBERRY (MORUS SPP.) CULTIVATION

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Abstract

Mulberry (Morus spp.) is the only food plant for the silkworm, Bombyx mori Linn., which is an economically important insect for sericulture industry. Mulberry is attacked by a number of insect pests, parasites, predators and pathogens around the year which not only affect the leaf quality but also responsible for poor yield. Mulberry is reported to be attacked by more than 200 species of insects belonging to various orders. Pests are of major significance for mulberry cultivation for foliage production. According to an estimate, the pests and diseases cause about 25% loss in foliage production of mulberry, besides deteriorating the nutritive value of leaves. Although several chemical pesticides have been suggested for the control of pests in mulberry these chemicals cannot be relied upon all the times due to their residual toxicity. Furthermore, since the chemical have to be applied repeatedly for the consistent pests and disease control, their unscientific usage can be hazardous to silkworm crop, besides polluting the environment and killing the beneficial microflora and fauna of the mulberry rhizosphere. Realizing the drawback of chemical measures of pest control, the usage of biocontrol agents has received maximum attention for the control of mulberry pests. The insect populations are naturally affected by a wide variety of environmental factors, both biotic and abiotic. The action of these factors is often termed natural control. One aspect of natural control is biological control, which involves the regulation of population of pests by the use of biotic agents. Biological control agents traditionally have been considered to be predators, parasitoids and microbial pathogens. Mulberry is subjected to damage by various species of sap suckers and defoliating insects. Among sap suckers mealybug, thrips, jassids, scale insect and white fly caused appreciable damage and play an important role in decreasing the crop yield significantly. The two major groups of defoliators that cause damage to mulberry leaves via their chewing mouthparts include the caterpillars and beetles. The present review focuses on the insect pests of mulberry and their management by using biocontrol agents in mulberry plantation.

Keywords: pests, parasitoids, crop loss, mulberry plantation

Introduction

Integrated pest management (IPM) is a broad ecological approach for managing pests by employing all methods and techniques viz., cultural, mechanical, chemical and biological. Crop pests constitute a major constraint to increase food production. Annual crop loss due to pests in India which at today prices would exceed Rs. 2,00,000 million per year. Since IPM is going to play an important role in Indian agriculture in the near future, overall efforts to develop economical and eco-friendly IPM techniques are needed (Kumar et al. 1994; Puri 1996). The important components of IPM to keep pest population below the economic injury level are the use of resistance varieties, conservation and augmentation of biological control agents, regular pest surveillance and use of sex pheromone for monitoring the pest build up (Puri 1996). The use of microorganisms in pest management has received greater attention in...
Recent years and commercial products like Bacillus thuringiensis (Bt) and nuclear polyhedrosis virus (NPV) are in use for control of Helicoverpa and Spodoptera. Mass multiplication of natural enemies and NPV are in progress at several places in the country. The losses caused by insect pests, diseases and nematodes in various agricultural crops account for 25-30 per cent.

Pests are of major significance for mulberry cultivation for foliage production. According to an estimate, the pests and diseases cause about 25% loss in foliage production of mulberry, besides deteriorating the nutritive value of leaves (Gupta et al. 2000). Although several chemical pesticides have been suggested for the control of pests in mulberry (Govindaiah et al. 2005), these chemicals cannot be relied upon all the times due to their residual toxicity. Furthermore, since the chemical have to be applied repeatedly for the consistent pests and disease control, their unscientific usage can be hazardous to silkworm crop, besides polluting the environment and killing the beneficial microflora and fauna of the mulberry rhizosphere. Realizing the drawbacks of chemical measures of pest control, the usage of biocontrol agents has received maximum attention for the control of mulberry pests. The insect populations are naturally affected by a wide variety of environmental factors, both biotic and abiotic. The action of these factors is often termed natural control. One aspect of natural control is biological control, which involves the regulation of population of pests by the use of biotic agents. Biological control agents traditionally have been considered to be predators, parasitoids and microbial pathogens.

1 Predators and Parasitoids: Insect predators and parasitoids play a very important role in biological control programmes. Biological control through insect predators involves three aspects viz., introduction, conservation, and augmentation. Introduction of exotic predators and parasitoids entails the search for natural enemies in other countries and their introduction into areas where the concerned pest insects are causing damage. Conservation of predators and parasitoids emphasizes the importance of preserving naturally occurring beneficial species (Luckman and Metcalf 1982). Conservation may involve either careful use of insecticides to avoid accidental mortality among beneficial groups or manipulation of the environment to make it more suitable for them, as by planting flowering plants to provide a source of nectar for adult parasitoids. Augmentation of predators and parasitoids involves the culture and repeated release of natural enemies to suppress pest populations. The beneficial insects may occur naturally, but at low densities, or they may be exotic species that do not persist in their new environment. Insects that are not pests sometimes attain pest status when beneficial insects (predators and parasites) are inadvertently killed by excessive insecticide use.

2. Pathogenic Biological Control: Control of insect pests by employing the use of microbes is equally eco-friendly due to their specificity to arthropods. So far, 3000 microorganisms have been registered to cause diseases to insects and a majority of them remain undescribed (Maddox 1994). A number of representative organisms from different types have been registered for use against the insect pests. The microbial control agents or pathogens are also important in naturally occurring biological control. The potential for pathogens in applied biological control probably exceeds that of many other pest management tactics, including predators and parasitoids. In many ways, pathogens are very similar to parasitoids. They are most effective against the immature stages of insects, and adults tend to be less susceptible or immune. As is often the case with parasitoids, they develop internally, and a single host can produce many pathogens. Pathogens often has a rather specific host relationship (narrow host range), much like parasitoids. However, active host "selection" is rarely involved; the pathogenic organisms are ingested inadvertently or spread by air, water, or oviposition by parasitoids. Acute, lethal infections are common, but chronic, debilitating infections also occur, whereas the attack of parasitoids is almost always fatal.
2.1 Viruses: Viruses are the acellular organisms and as usual, particles of many insect viruses are enclosed in protein crystals called inclusion bodies (Figs 1-2). The principal types of inclusion viroses are Nuclear Polyhedrosis Virus (NPV), Granulosis Virus (GV), Cytoplasmic Polyhedrosis Virus (CPV), and entomopox virus. Non-inclusion viroses occur in insects but are difficult to detect without an electron microscope and are not well-known. NPVs multiply in the cell nucleus of the host tissues (Fig. 3). Tissues most commonly infected are the epidermis, fat body, blood cells, and tracheae in Lepidoptera (Tewari et al. 1996). In phytophagous Hymenoptera, midgut cells are infected. The GV's multiply in both the nucleus and the cytoplasm of host cells. The fat body is the principal site of infection, but the epidermis and tracheae are sometimes infected. Symptoms of infection are similar to NPV infection. Lepidoptera are common hosts. CPV's develop in the cytoplasm of the host cells. The midgut is infected, and Lepidoptera larvae are the usual hosts. The disease tends to be debilitating, diseased larvae exhibit loss of appetite, are smaller, and develop slowly. Insects that are infected but not killed may have a reduced reproductive capacity. CPV’s are not as host specific as are the other two virus groups mentioned previously. Viruses seem to play a significant role in the pest management.

2.2 Bacteria: Bacteria are prokaryotic organisms and the important bacterial pathogens of insects form resistant spores that can survive in unfavorable conditions quite well. Non-spore-forming bacteria are common in the digestive tracts of insects, but they are pathogenic only when they invade the hemocoel, this occurs under unusual conditions such as stress or injury. The best-known insect pathogens among the spore-forming bacteria include Bacillus thuringiensis and B. popiiliae. B. thuringiensis, was first isolated from silkworm larvae by Japanese workers about 1900. There has since been extensive research on this organism, and several strains infecting a wide variety of insects are recognized, however, it is most frequently applied against lepidopterous larvae. It has been pointed out that the organism was easy to produce in large quantities and to disseminate as a "microbial insecticide." Infected caterpillars become sluggish and eventually discolored and flaccid. Hundreds of tons of B. thuringiensis spores are now being produced commercially each year, under such names as "Thuricide," "Dipel," and "Biotrol". The bacterial cells contain a toxic protein crystal as well as the spore. When dissolved in the insect, the crystal causes paralysis of the gut. However, both spore and crystal are usually required for effective action. Varatharajan et al. (1996) described the effect of B. thuringiensis var Kurstaki on the caterpillars of Spilosoma obliqua and Pieris brassicae.

2.3 Fungi: Fungi are important naturally occurring biological control agents. There are a wide variety of fungi that are insect pathogens. Entomopathogenic fungi often are not very specific. Fungal pathogens are usually transmitted from host to host by spores; unlike viruses and bacteria, fungi are capable of penetrating the insect directly through the integument (Kumar et al. 1997a; Kumar et al. 1999b; Kumar et al. 2004). Fungi, more than any other insect pathogen, require favorable environmental conditions for development of epizootics, or outbreak of disease. This seems to be due to the need for high humidity that usually is required for germination of fungal spores. Fungi can often be cultured on artificial media, but infectivity is reduced. Some species have been used successfully to reduce pest populations, Verticillium lecanii is used for biocontrol of aphids and scales affecting greenhouse/crops, Beauveria bassiana is applied against a variety of pests (Reithinger et al. 1997) and Metarhizium anisoplea is used for controlling spittlebugs on sugarcane. Similarly, Hirsutella thompsoni is used for suppression of citrus rust mites.

2.4 Protozoa: These are the microscopic a cellular organisms faming a heterogeneous assemblage of organisms. Many groups, including the flagellates, gregarines, and amoebae, are pathogenic to insects. The order Microsporida, especially pathogens of the genus Nosema, has received considerable attention by entomologists. Microsporidians can be transmitted orally or via eggs (Tanaka et al. 1972; Fujiwara 1980). They are not very specific and often cause a chronic disease that result in lowered fecundity or
sterility. Nosema locustae affects a large number of grasshoppers and can be produced in commercial quantities by inoculating immature grasshoppers in culture. One infected grasshopper will normally produce enough spores to treat 3 to 4 acres of rangeland. Nosema spores usually are mixed with bran to make bait that grasshoppers ingest readily. Nosema also can be applied in conjunction with a reduced amount of insecticide to provide rapid, but residual, suppression of grasshopper populations.

Biocontrol Agents in Mulberry Pest Control

Mulberry (Morus spp.) is the only food plant for the silkworm, Bombyx mori Linn., (Fig. 4) which is an economically important insect for sericulture industry. Mulberry is attacked by a number of insect pests, parasites, predators and pathogens around the year which not only affect the leaf quality but also responsible for poor yield (Kumar 1999; Kumar et al. 2001; Kumar 2002a). Mulberry is reported to be attacked by more than 200 species of insects belonging to various orders (Zheng et al. 1988). The total loss of mulberry foliage crop due to these natural enemies is about 20% per year (Gupta et al. 2000). The present review focuses on the insect pests of mulberry and their management by using biocontrol agents in mulberry plantation.

Mulberry is subjected to damage by various species of sap suckers and defoliating insects. Among sap suckers mealybug, thrips, jassid, scale insect and white fly caused appreciable damage and play an important role in decreasing the crop yield significantly. The two major groups of defoliators that cause damage to mulberry leaves via their chewing mouthparts include the caterpillars and beetles.

1 Mulberry Sap suckers: About 30 species of sap suckers were observed to inflict damage on mulberry leaves which include 18 heteropterans, 10 homopteran and 2 thysanopteran species. Further, sap suckers of mulberry were categorized into major pests (2 species), minor pests (15 species) and occasional pests (13 species).

11 Mealybug, Maconellicoccus hirsutus (Green): The mealybug, M. hirsutus (Green) has attained a major pest status in mulberry and also popularly known as “hard to kill” pest. It is also called as “Pink hibiscus mealybug” and Grapevine mealybug. The mealybug has been recorded to infest more than 300 plant species. In mulberry, it causes a disease called as “tukra” Katiyar et al. 2001a. The average incidence of tukra in the Southern states of India is recorded to be 34.24% (Manjunath et al. 1996) and the reduction in leaf yield is estimated to be 4500 kg/ha/yr (Kumar et al. 1994). The typical symptoms of tukra are wrinkling of apical leaves, swelling of the shoot and shortening of internodal distance (Fig. 5). In case of heavy infestation, the leaves on the apical shoot turn yellow and shed prematurely. Finally, the tukra affected plants acquire stunned growth leading to reduction in leaf yield (Katiyar et al. 2001a). Mealybug infects the apical tender leaves of the plant which are used for the chawki (early stage larvae) rearing of silkworm. As it feeds, the mealybug injects into the plant toxic saliva that results in malformed leaf and shoot, stunting and occasional death of the plant. Infected apical leaves show a characteristic curling. Mealybug have long, thread like mouthparts (styles) which penetrate plant tissues with the bristle like mandibles and maxillary styles and suck the cell sap of young stem and leaves. The mealybug also reported as vector for many plant diseases. Tukra was earlier believed to be a viral disease transmitted through the mealybug (Babu et al. 1994). But the more recent experiments conducted at the Central Sericultural Research and Training Institute, Mysore, (Manjunath et al. 1996) ruled out the involvement of virus, and tukra is now considered as a manifestation of mealybug attack, even though the precise genesis of the symptom is still not understood (Babu et al. 2004). Because of its typical habitat i.e., under the crumpled leaves, and at the base of the leaf petioles together with covering of egg masses, and the covering of late age nymphs and adults in their waxy filamentous secretions...
(Figs. 6-7) secreted through the dermal pores (Figs. 8-9) (Kumar et al. 1997b) protected the mealybug are themselves against exposure to their natural enemies and insecticides. Thus insecticidal applications are not very effective against the mealybug population. Therefore, biological control gained importance to control the mealybug in sericulture industry. The biocontrol agents found effective against the mealybug are:

1.1 Lady Bird Beetle, Cryptolaemus montrouzieri Mulsant: Commonly known as lady bird beetle, M. montrouzieri has shown immense potential in management of mealybug infesting mulberry plantation. It feeds on the larval and adult stage of mealybug (Fig. 10). The adults of the beetle are to be released in the field @ 200-300 adult females per acre of mealybug infested mulberry gardens. It can easily trace the sites of mealybug hiding on the mulberry plant where chemical insecticide spray does not reach. Following reports on the successful management of the pink mealybug with an exotic predator, C. montrouzieri Mulsant in grape vineyards (Sysoev 1953; Ranga Reddy and Lakshmi Narayana 1986; Mani and Thontadarya 1988) and tobacco plants (Gautam et al. 1988), the release and evaluation of the predator has been considered for managing the mealybug in mulberry. Chakraborthy et al. (1996) described the field efficacy of exotic predator C. montrouzieri on mealybug infested mulberry. The predators which devour all life stages of mealybug by ovipositing its eggs in the mealybug colony. In the first week of their study the grubs of C. montrouzieri started to appear (0.425/tukra infested shoot) and then gradually increased during the following three weeks as 0.85, 0.90 0.95/tukra infested plant. On the 35th day of the release of the predator, the grub population was found to be decreased considerably as 0.3, which further reduced to 0.05 on the 42nd day. The reasons for decline in the grub population of C. montrouzieri were due to declining of the prey population and cannibalism amongst the grubs when food became scarce. Further, they concluded that the predator @ 5 adults/plant suppressed the mealybug population effectively in 21 days and reduced to nil in 42 days. They also recommended that inoculative releases of C. montrouzieri in mulberry fields may help the predator’s establishment and control of M. hirsutus.

1.1.2 Scymnus nubilus Mulsant: Another coccinellid beetle, S. nubilus Mulsant was also reported as a predator of pink mealybug, M. hirsutus. Both grubs and adults of S. nubilus were found to feed on all stages of the host. Each grub of S. nubilus consumed 1,320 eggs or 208 nymphs or 28 adults whereas, an adult predator devoured 2301 eggs or 326 nymphs or 74 adults of mealybug. It was estimated that during the entire feeding period an individual predator could prey 3622 eggs or 534 nymphs or 103 adults of mealybug (Santha Kumar et al. 1996). Even though C. montrouzieri is reported to have a better feeding efficacy over S. nubilus (Mani and Thontadarya 1967, 1988), C. montrouzieri is sensitive to both low and high temperature and can survive only within a range of 20-30 °C temperature. Hence, its multiplication in the field, where the temperature is as low as 5 °C in winter, and as high as 45 °C in summer, is not satisfactory throughout the year. But, S. nubilus is a well adapted native predator and has got a better chance of its utility for biological control of M. hirsutus (Santha Kumar et al. 1996).

1.1.3 Anagyrus kamali (Moursi): Parasitization of mealybug by A. kamali was reported earlier and Egypt was reported by Moursi (1948). Noyes and Hayat (1984) listed A. kamali from India as Anagyrous sp. Recently, Sagarrar and Vincent (1999) presented a brief account on the biology of the parasitoid on M hirsutus. The occurrence of A. kamali, a solitary endoparasite of the mealybug, M. hirsutus has been reported for the first time from India (Katiyar et al. 2000). The parasitoid was found to be parasitizing the field population of mealybug to the tune of 10.37 to 42.70% in different months (Figs. 11-14). The development of the parasitoid on different stages of mealybug indicated that the parasitoid was able to complete its development in all the stages. Higher parasitism (67.48-78.08%) and more female progeny
were observed when third instar nymphs and adult female of the mealybug were exposed to the parasitoid. The parasitoid completes its life cycle in 19.72±1.12 days. On an average each female of A. kamali laid 39.0±4.53 eggs. It was found to parasitizing 8-10 mealybugs and depositing 1-3 eggs per host individual (Katiyar et al. 2000).

2. Mulberry Defoliators: Defoliating insects are common in all agricultural ecosystems. They are detrimental to the health and productivity of plants. Depending upon the duration and the severity of defoliation, plant growth may be negatively affected. Early detection and identification of an outbreak and identification of the causal insect are critical initial steps in managing defoliators. Signs of insect defoliators include:

- Defoliation
- Presence of insect life stages
- The presence of frass on ground or on leaves
- Silk webbing, or streamers on ground or in foliage

The major groups of defoliator that cause damage to plants via their biting and chewing mouthparts are various species of caterpillars. In addition to caterpillars, others chewing insects that are sometimes damaging to mulberry include beetles, grasshoppers, katydids and their relatives. Two major defoliators cause extensive damage to mulberry plantation in sericulture industry viz., Bihar hairy caterpillar (Spilarctia obliqua, Walker), and leaf roller (Diaphania pulverulentalis, Hampson) and a minor defoliator cutworm, Spodoptera litura.

2.1 Bihar Hairy Caterpillar (Spilarctia obliqua Walker): S. obliqua commonly known as Bihar hairy caterpillar is one of the major pests of mulberry, which usually infest mulberry crop from July to February in Kamataka (Ramkishore et al. 1994; Shree and Manjunatha 1998; Kumar et al. 1999b). A total foliage crop loss in mulberry by this pest was estimated around 4.90% during 1996-1997 (Shree and Manjunatha 1998). The larval infestation is sporadic and it is potentially destructive to a wide variety of vegetables, pulses, oilseeds and even some medicinal plants (Ahmed and Kumar 1993). Sharma and Tara (1988) reported that an individual larva of S. obliqua consumes 1195.18 g of mulberry leaves in its entire larval period. Several chemical and mechanical control measures have been developed and adopted to eradicate this polyphagous pest. Still, SoNPV continues to be the most destructive insect pest for mulberry and many other agricultural crops.

In addition to the chemical insecticidal control the management of the various developmental stages viz., egg, caterpillars and pupae of S. obliqua can be managed by using biocontrol agents. The eggs of S. obliqua can be successfully parasitized by a hymenopteran wasp, Trichogramma chilonis. While three early instars of S. obliqua in various agricultural crops is based on population monitoring by its two natural parasitoids viz., Apantalis (Kalara 1984; Katiyar and Sharma 1987; Muthukrishnan and Senthamizhvel 1987; Shetgar et al. 1990) and Meteorus (Kumar and Yadav 1987), the later instars of the pest in mulberry plantation can be controlled by a predatory bug, Eocanthecona furcellata (Kumar et al. 2001). The biocontrol agents found effective against the Bihar hairy caterpillar are:

2.1.1 Egg Parasitoid (Trichogramma chilonis): The egg parasitoid, T. chilonis has been found to be a potential biocontrol agent against Bihar hairy caterpillar in the mulberry plantation (Fig. 15). Release of T. chilonis in mulberry crop @ 1 lakh adults/acre at an internal of 3 days, parasitized 33.12% eggs of Bihar hairy caterpillars (Katiyar et al. 1999; Govindaiah et al. 2005). T. chilonis was found to destroy an appreciable population of S. obliqua eggs upto 5 meter from release point although it was able to search and parasitize the host eggs upto 10 meters. T. chilonis has been exploited as a biocontrol agent and its
field efficacy has been tested against several agricultural pests. The rate of parasitization in other pest by T. chilonis ranged from 12.6 to 90% (Katiyar et al. 1999). Takada et al. (2000) reported that the egg parasitoid, Trichogramma dendrolimi grew rapidly on the eggs of Mamestra brassicae to full size by second day after parasitism, sucking up nearly all of the host yolk as the food resource. The emergence of adult weaps from the host egg occurred ninth day after parasitization. Further, the female of T. dendrolimi was able to parasitized M. brassicae eggs of all ages.

2.1.2 Meteorus dichomerides: M. dichomerides (Willison) (Hymenoptera : Braconidae) is an indigenous, natural solitary, potential biocontrol agent of Spilarctia obliqua Wilk. (Fig. 6). The mated females preferred the early two instars of its host, since these stages are gregarious in nature and parasitoid found easy to lay the eggs in a single colony of the host without much resistance (Katiyar et al. 2001a). Once the host is fixed, the parasitoid bent its abdomen downwards and injects its ovipositor into the host body to lay an egg (Fig. 7). On an average a single mated female lays 22±0.98 eggs in its life span. The maggots of M. dichomeridis moult three times during the total life span of 11.95±0.45 days (Kumar et al. 2005). The first two moult occurred inside the host while the third moult took place out side of its host body. After attaining full growth, the mature maggot stopped feeding and cut a hole in the dorsolateral region of its host body and emerged out (Fig. 8). After the emergence, it searched for a suitable place for pupation and started spinning a brown silken cocoon, within which it pupated (Fig. 19). M. dichomeridis completed its life cycle in 19.24±0.79 days. The longevity of male and female was observed as 46.20±0.52 and 49.20±0.52 days, respectively. M. dichomeridis substantially parasitized field population of S. obliqua (ca. 25%) suggest its potential agent for S. obliqua (Katiyar et al. 2001b).

2.1.3 Eocanthecona furcellata: The stink bug, E. furcellata (Wolff.) is a natural and potential biocontrol agent of Spilarctia obliqua (Kumar et al. 2002b). On locating the moving prey by its visual stimuli the predator raises its body from the leaf surface and moves towards the prey with continuous antennal movements (Kumar et al. 2001). If the prey is larger in size (IV and V instar larvae), the predator jabs the prey repeatedly with the fully extended rostrum and injects saliva (Fig. 20). The predator, probing three or four times to select its feeding site, holds the host plant leaf by its legs and lean forward towards the prey for stylet penetration. Feeding commences immediately after killing or immobilizing the prey (Fig. 21). If the prey is small, after immobilizing it, the predator feeds on it without changing its feeding site till the body fluid is completely desapped (Fig. 22). The feeding is continued till the predator is satiated and further predation commences only after 56 hrs. SEM microphotographs revealed that the rostral tip jabs at the posteroventral region of the prey particularly, in between the abdominal legs. The stylet bundles are seen protruding from the rostral tip and penetrating into the prey body (Fig. 23). The adult female predator is more voracious feeder than the adult male and consumed 41.9±0.64 small larvae and 42.2±0.87 large larvae during their life span. The longevity of male and female was observed as 20.7 and 29.4 days respectively. Visualization of the predator as well as the movement of the prey increases the predatory efficiency. Field observations indicated a drastic fall in the incidence of the mulberry pest, S. obliqua with the increased population of E. furcellata in mulberry plantation (Kumar et al. 2001).

2.1.4 Spilarctia obliqua Nuclear Polyhedrosis Virus (SoNPV): While surveying the mulberry germplasm of Central Sericultural Research and Training Institute, Mysore, for the incidence of pests, numerous larvae of S. obliqua were found diseased or dead and hanging from the twigs (Fig. 24) of mulberry plants (Kumar et al., 2000). Polyhedral occlusion bodies were isolated from the hemolymph of dead larvae. The scanning and transmission electron microscopic examination of the inclusion bodies confirmed the pathogen as nucleopolyhedrosis virus (Kumar et al. 2000). On inoculation, SoNPV was
found to be highly pathogenic to fifth instar larvae of *S. obliqua*. The dosage-mortality line indicates that the larval mortality was directly related to number of POBs administered orally to *S. obliqua*. LC$_{50}$ value for fifth instar larvae was $0.1485 \times 10^4$ POBs/ml. The inoculated larvae showed similar symptoms like the field infected larvae. The larvae succumbed to death within 6-7 days after inoculation in laboratory conditions. Under field condition, SoNPV were shown to be highly effective biopesticide against *S. obliqua*, which infests mulberry. Battu (1982) and Battu and Ramakrishnan (1987) revealed that the nuclear polyhedrosis virus was found almost continuously associated with *Diacrisia* (= *Spilarctia*) *obliqua*. Further, Battu and Ramakrishna (1987) observed that the 6-day old larvae of *D. obliqua* were most susceptible among the age group tested (Kumar et al. 2000). The symptoms of the NPV infected *S. obliqua* generally resembled that described for other NPV infected lepidopterous larvae (Ignoffo and Garcia 1979; Battu 1982; Johnson and Lewis 1982; Tewari et al. 1996; Entwistle 1998; Moscardi 1999). In the early phase of the disease no symptoms were noticed but with the progress of the disease the diseased larvae of *S. obliqua* were found slightly sluggish and lagged behind in their development. SoNPV appears to have great potential as a microbial control agent for this insect pest than other microbial insecticide investigated to date.

### 2.1.5 Spilarctia obliqua Cytoplasmic Polyhedrosis Virus (SoCPV)

The cytoplasmic polyhedrosis virus was isolated from a few dead caterpillars of *S. obliqua* on field beans. The diseased caterpillars were small and on tissue examination revealed numerous inclusion bodies (Narayanan 1986). The symptoms of the CPV infected *S. obliqua* generally resembled those that described for other CPV infected lepidopterous larvae (Smith 1963). Living diseased larvae of *S. obliqua* showed typical sluggishness in their movement and they lagged behind in their development and they were also less responsive to tactile stimuli.

### 2.2 Leaf Roller, *Diaphania pulverulentalis* (Hampson):

In recent years, a new invader, leaf roller, *D. pulverulentalis*, is becoming a growing threat to mulberry in Southern states of India and the average incidence by this pest was recorded 27.85% in Karnataka, 20.98% in Andhra Pradesh and 16.48% in Tamil Nadu states which alarmed to take appropriate control measure to suppress it population (Kumar 1999; Rajadurai et al. 1999). Generally, the leaf roller appears during June and persists upto February.

*D. pulverulentalis* infests apical portion of mulberry shoot by binding of leaflets together with silky secretion facilitating the larva to settle inside and devour the leaf content. The infected leaves are not suitable for mulberry silkworm rearing. The life cycle of *D. pulverulentalis* is comparatively shorter than other prevailing pests in mulberry plantation which makes the pest to proliferate rapidly with more number of generations in a year. It completes life cycle in 17-24 days. Mated female lays eggs singly along midrib of mulberry leaf ([Fig. 25](#)) which hatched out within 2-3 days. In view of leaf yield loss caused (12.13%) and the infestation reaching the economic injury level during the peak period, it is imperative to suggest the prophylactic control measures before the multiplication of the leaf roller population in the mulberry plantation. Recently, Rajadurai et al. (2003) studied the impact of infestation of leaf roller on growth and yield of mulberry. The biocontrol agents found effective against the leaf roller are:

#### 2.2.1 *Trichogramma chilonis* (Hymenoptera: Trichogrammatidae):

* T. chilonis wasp has been utilized more than any other natural enemy for field release to control lepidopteran pests of various agricultural corps including mulberry pests. *T. chilonis* is a small tiny egg parasitoid parasitizes the eggs ([Fig. 26](#)) of leaf roller upto 80.33% (Govindaiah et al. 2005). The parasitoid female oviposite in the host eggs which turn black after three days of parasitization. The parasitoid emerges out from the leaf roller egg after 8-9 days killing the egg.
2.2.2 *Tetrastichus howardi*: The indigenous gregarious hymenopteran parasitoid was found to parasitize (Fig. 27) the pupae of leaf roller in the laboratory. The incubation period was observed 13-15 days and adult male and female longevity extends up to 14 and 17 days respectively. The parasitization rate of 75.67% was recorded from the laboratory population (Rajadurai et al. 1999).

2.3 Cutworm. *Spodoptera litura*: The cutworm, *S. litura* is a leaf feeder of mulberry and was found predating by a sting bug.

2.3.1 *Eocanthecona furcellata*: Sting bug, *E. furcellata* was found predating cutworm, *Spodoptera litura* during the pest and survey in mulberry plantation (Fig. 28) of Central Sericultural Research and Training institute, Mysore, (Govindaiah et al. 2005). Various natural enemies available in the field are fair enough for utilization in the biocontrol programmes against the major and minor pests of mulberry.

Fig. 1. Scanning electron micrograph of BmNPV polyhedra (P).

Fig. 2. After alkali treatment a disintegrated polyhedral body (P) of BmNPV showing a large number of released virus particles (V).

Fig. 3. Transmission electron microphotograph reveals the infected midgut nucleus (N) with two polyhedra (P) of BmNPV (Nm = Nuclear membrane).

Fig. 4. Last instar silkworm, *Bombyx mori* larvae (Sl) feeding on mulberry leaves (Ml).

Fig. 5. Mulberry twig infested with mealybug, *Maconellicoccus hirsutus* (arrows), (Tal = Tukra affected leaves).

Fig. 6. A view of the mealybug ovisac (Os) showing eggs (E), nymphal instars (Ny) covered with secreted wax (Wx).
Fig. 7. Dorsal view of the first instar of mealybug.

Fig. 8. Scanning electron microphotographs of posteroventral view of adult female of mealybug showing tubular ducts (Td), trilocular pores (Tp) and multilocular disc pores (Mp).

Fig. 9. A multilocular disc pore (Mp) of adult female of mealybug reveals the extruding spiral wax (Wm).

Fig. 10. Coccinellid beetle (Bt), C. montrouzieri feeding on different stages of mealybug (Mb).

Figs. 11-12. Adult female of Anagyrus kamali on female nymphal body of mealybug.

Fig. 13. Parasitized mealybug. Arrows indicate the sites of parasitization by A. kamali.

Fig. 14. Mummified mealybug.

Fig. 15. Adult female of Trichogramma chilonis (arrow) parasitizing eggs (Eg) of Spilarctia obliqua.
Fig. 16. A gravid female of Meteorus dichomeridis with stretched antenna (An).

Fig. 17. Gravid female of M. dichomeridis bents its abdomen and penetrates its sharp ovipositor (Op) into the host body to facilitate oviposition (Hl = Host larva).

Fig. 18. A fully grown third instar maggot (Mg) of M. dichomeridis emerges from the dorsolateral region of the posterior end of its host larva (Hl) of S. obliqua.

Fig. 19. The host larvae (Hl) of S. obliqua from which the maggot of M. dichomeridis emerged, and the maggot turned into a pupa (Pu).

Fig. 20. Predatory bug (Pd) of Eocanthecona furcellata feeding on S. obliqua larvae (Hl). Arrow indicates the feeding site. (Rs = Rostrum).

Fig. 21. E. furcellata holding its prey at the rostral tip and moving at the leaf margin and desapping the body fluid of the prey.

Fig. 22. Predatory bug (Pd) almost desapped the body fluid of its host larva (Hl).
Fig. 23. Scanning electron micrograph showing the stylet (St) penetration (arrow heads) of the predator, E. furcellata into the host larva body (Hlb) at the posteroventral region. (Rs = Rostrum).

Fig. 24. Dead last stage larva of Spilarctia obliqua by SoNPV infection. Droplets on the lower leaf surface are of hemolymph oozed out (circles) from the larval body.

Fig. 25. SEM microphotograph reveals a single egg (Eg) encircled by leaf trichome (Ltr) and attached between the leaf veins on the adaxial surface of mulberry leaf.

Fig. 26. Adult female of Trichogramma chilonis (Tc) parasitizing the eggs (arrow heads) of leaf roller, Diaphania pulverulentalis.

Fig. 27. Adults of Tetrastichus howardii (Th).

Fig. 28. Late age nymph of Eocanthecona furcellata feeding on its host larva (Hl) of Spodoptera litura. (Pd = Predator; Rs = Rostrum).

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