Biological Control of Insect Pests in Mulberry Sericulture

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ISO 9001: 2008 Certified

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Month and Year of Publishing: January 2016

Language: English

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Layout
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Photo courtesy
NBAIR, Bengaluru
A. M. Babu
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Published by
Director
Central Sericultural Research & Training Institute (CSRTI)
Central Silk Board (CSB), Ministry of Textiles: Govt. of India
Manandavadi Road, Srirampura, Mysuru-570 008, Karnataka
www.csrtimys.res.in

Foreword

Mulberry (*Morus* sp.) is a perennial, deep rooted, high bio-mass producing and protein rich foliage plant; the sole food plant of mulberry silkworm (*Bombyx mori* L.). It also forms an unlimited source of food and shelter for a variety of pests, which cause considerable damage to foliage in varying intensities. Although more than 300 species of pests (insect & non-insect) have been reported to infest mulberry, a few soft bodied insects such as pink mealybug, papaya mealybug, thrips, whitefly etc. and defoliators such as leaf roller & Bihar hairy caterpillar are considered as major pests causing significant qualitative and quantitative damage. Uzi fly (*Exorista bombycis* Louis), a dipteran tachinid fly infestation causes crop losses in silkworm cocoon crop.

The negative impact of pesticides on human health and the environment is leading to renewed interest in developing biological pest control methods as alternative for sustainable crop yields. Further, there is little scope for using chemicals towards crop protection in sericulture, owing to harmful effects of pesticides on silkworms. Moreover, tremendous advances have been made in beneficial organism technology, especially insect predators and parasitoids. However, developing techniques to mass produce these biological control agents is very difficult and not enough as they should be effective and economically feasible.

The success of biological control by releases of natural enemies largely depends on our ability to mass-produce the required biological control agents for timely releases. In this context, bringing out a technical bulletin documenting essential information about the bio-control agents and the protocol for their mass multiplication would benefit of crop protection and extension personnel besides stakeholders in sericulture. The bulletin would also serves as a guide to unemployed rural youths for establishing insectary and take up mass production of bio-control agents as an entrepreneurial activity.

V. Sivaprasad
Director, CSRTI-Mysuru

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Introduction

The successful management of a pest by means of another living organism/natural enemy that is encouraged and disseminated is called as biological control. In such programme, the natural enemies are introduced, encouraged, and multiplied by artificial means and disseminated by humans on his own efforts instead of leaving it to the nature. The natural enemy may be a parasite/parasitoid, predator, pathogen etc.

Parasite

- An organism that lives at the expense of another (host), which it does not necessarily kill
- Usually smaller than the host
- Can't live independently and obtain nourishment from the host
- Relationship lasts till the life time of the host

Parasitoid

- A parasitoid is an organism that spends a significant portion of its life history attached to or within a single host organism
- It ultimately kills and consumes its host in the process
- The host usually dies before it can reproduce
- Usually, the term parasitoid refers only to insects with this type of life history

Predator

- An organism that kills & eats more than one other organism during its life

In recent years, there is an increasing awareness among the pest control specialists that parasitoids, predators and pathogens have immense potential to keep the insect pest populations in check. Among them, parasitoids have highest potential with 82% success rate in comparison to predators (17%) and pathogens (1%).

Why biological control?

Chemical control pose serious unwanted problems like air, soil &

water pollution, health hazards, killing of beneficial insects, secondary pest out-break, pest resurgence, pest-resistance etc. Therefore, biological control serves as a potential weapon in avoiding these problems.

Advantages of biological control

- Pollution free and non-hazardous
- Safe to handle and free of side effects
- Biological agent survives as long as the pest is prevalent and hence, control is effective over longer periods
- Pest is hunted out and thus complete control over a large area is possible
- Solves the problem of pests by perpetuating itself
- Safe to non-target organisms
- Compatible with other methods

Disadvantages of biological control

- Slow process and takes longer time
- Natural enemies cannot be restricted to a particular pest, crop or area
- Not applicable to all the species of pests
- Presence of alternate hosts delays the biological control
- If hyper parasites are there, the effect of parasites is adversely affected
- Bio-agents have less shelf-life
- Non-availability of bio-control agents**Biological control of major sericultural pests**

Both mulberry and silkworm crops are prone to attack by several species of insect pests in varying intensities. Mulberry pests cause 10-15% leaf yield loss. Major mulberry pests are briefed below:

Tukra mealybug, (*Maconellicoccus hirsutus*)

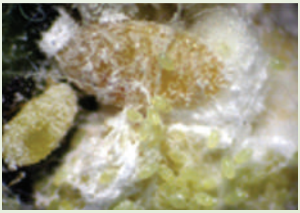
Also known as pink hibiscus mealybug or grapevine mealybug and are regarded as hard to kill pests. They are minute, soft bodied and polyphagous (>300 alternate hosts). They undergo parthenogenesis. Wax coating over the body surface and ovisac provides protection against contact insecticides. They are resistant to systemic insecticides. They occur throughout the year, while the incidence is high in summer. They are estimated to cause a leaf yield loss to an extent of 4500 kg/ha/yr.

Damage symptoms in mulberry

- White deposits of filamentous waxy secretion on the affected stem / branches
- Wrinkled, thickened, brittle & dark green leaves on apical portion of shoot
- Leaves breakup into pieces on pressing, hence called as Tukra (synonymous to Hindi word tukda)
- Shortened internodes results in 'bunchy top' appearance
- Distorted apical portions of twigs

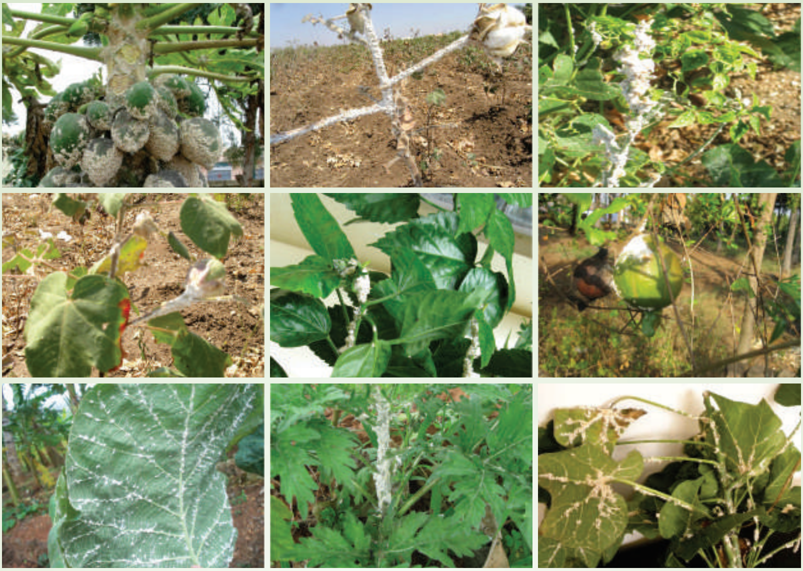
Papaya mealy bug, *Paracoccus marginatus*

Exotic pest (native to Central America) and believed to have arrived to India from Sri Lanka. First reported on mulberry in Tamil Nadu (January 2009). Subsequently spread to all the southern states and Maharashtra. Occurs throughout the year and the incidence is high during summer months.



Alternate host plants

Papaya is the basic host plant of papaya mealybug. However, it has wider host range (>250) including cotton, cowpea, brinjal, *Hibiscus*, pomegranate, teak, *Parthenium*, *Jatropha*, guava, beans, *Plumeria*, *Abutilon*, *etc.*, besides mulberry.



Damage symptoms in mulberry

- Attacks leaf buds, leaves, stem, fruits *etc*.
- Affected leaves turn yellow and plant growth retards
- Deformation of plant parts especially leaves and growth of sooty mould
- Heavy infestation leads to drying and death
- Attack on emerging eye buds on pruned stumps causes death of whole plantation

Mulberry leaf roller, *Diaphania pulverulentalis*

Major seasonal pest in the southern states and infestation coincides with the onset of monsoon. Peak period of infestation is from September-December. Infestation is observed from 15 days through 70 days after pruning. It causes considerable reduction in leaf yield.

Damage symptoms in mulberry

- Target area of attack is the apical portion, thus affecting chawki leaves
- Young caterpillar binds the leaflets together by silky secretion, settles inside and devours the soft green tissues of the leaf surface
- Grown up caterpillars feed on tender leaves
- Faecal matter of pest can be seen on the leaves below the affected portions

Bihar hairy caterpillar, *Spilosoma obliqua*

Polyphagous pest with alternate hosts like sunflower, safflower, green gram, jute, castor, teak etc. Voracious leaf eater causing extensive damage to mulberry. Occurs during monsoon and post monsoon periods. Severe infestation is reported from August to February.

Damage symptoms in mulberry

- Feeding of young age gregarious caterpillars lead to dried/mesh like appearance of leaves
- Grown up caterpillars feed on entire leaf resulting in branches without leaves

Silkworm uzi fly, *Exorista bombycis*

Uzi fly is a serious pest of silkworm inflicting 10-15% loss to silkworm cocoon crop. Infestation prevails all through the year, but high during rainy season followed by winter & summer.

Symptoms

- Uzi fly lays one or two cream colored eggs on the silkworm body
- Generally prefers late age silkworms for egg laying and eggs hatch in about 36 to 48 hours
- Black scar is formed at the point, where the maggot bores into the body of the silkworm is the pointer to the pest attack
- Uzi maggots emerge from the cocoon resulting in a circular exit hole at the tip, if the infestation is during V instar



Relevance of Biocontrol Agents

In sericulture, application of insecticides is not advisable because of toxicity to silkworms, environmental pollution and resurgence of pests due to elimination of natural enemies. Therefore, biological control, which is non-chemical & eco-friendly is the ideal alternative. Following bio-control agents are recommended by CSRTI-Mysuru to obviate the loss caused by major sericultural pests.

- *Cryptolaemus montrouzieri* and *Scymnus coccivora* (lady bird beetles) against pink mealybug
- *Acerophagus papayae* against papaya mealybug
- *Trichogramma chilonis* against lepidopteron pests
- *Nesolynx thymus* against uzi fly

Lady bird beetles against pink mealybug

Lady bird beetles are small, hemispherical beetles with well defined spots or other colored markings on the wings cases. Both adults as well as grubs (larvae) feed voraciously on all the stages of mealybugs, scale insects and other soft bodied insects. They often wipe out entire colony of pests. Although several species of lady bird beetles occur in mulberry gardens, *Cryptolaemus montrouzieri* and *Scymnus coccivora* are found to be effective against pink mealybug.

Cryptolaemus montrouzieri

Adult female lays pinkish eggs either singly or in groups of 2 to 6 embedded in the mealybug colonies generally in the late evening to early morning hours (between 10 PM to 6.00 AM). During day time oviposition is extremely rare.

Life cycle

Fecundity	:	150 - 200
Incubation period	:	5 - 6 days
Larval (grub) period	:	15 - 17 days
Pre-pupa	:	2 - 4 days
Pupal period	:	7 - 9 days
Total deve. period	:	29 - 36 days
Pre-oviposition period	:	6 - 8 days
Oviposition period	:	25-30 days
Adult longevity	:	Male 21 - 48 & Female 20 - 65 days

Adult emergence takes place normally in the morning hours (8.00 AM – 11.00 AM) and rarely in the afternoon. There is no emergence during night. Adult spends 1-2 days within the pupal case before emergence by breaking the case ventrally.

Predatory potential: Grubs are more voracious than adults and consume about 900 – 1500 eggs or 300 nymphs or 30 adult female mealybugs. Similarly an adult consumes 200 eggs or 50 crawlers.

Host range: Whitefly (*Aleurodicus dispersus*), aphids, Psyllids, scale insects, thrips (*Scirtothrips citri*) and few caterpillar pests.

Scymnus coccivora

They are potent and indigenous predators of wide range of mealybugs, aphids and scale insects during different stages of their development. Adults are light yellowish with dark brown markings along the mid-dorsal regions.

Life cycle

Fecundity	:	40-60
Incubation period	:	4-5 days
Larval (grub) stage	:	12-15 days
Pre-pupa	:	1-2 days
Pupal period	:	6-8 days
Total develop. period	:	23-30 days
Pre-oviposition period	:	6 - 8 days
Oviposition period	:	8-10 days
Adult longevity	:	Male 61 & female 69 days

Predatory potential: Grubs consume about 300 eggs or 65 nymphs or 7 adult female mealybugs during its life time.

Host range: Mealybugs, scale insects & other soft bodied insects.

Schedule & Method of release

It is recommended to release 250 *Cryptolaemus montrouzieri* or 500 *Scymnus coccivora* beetles /acre/year in 2 split doses at an interval of 6 months. Open the lid and release 2-3 beetles on the shoots having tukra symptom.

Acerophagus papayae against papaya mealybug

They are exotic, host specific, early nymphal parasitoids which are very minute in size. They actively search for papaya mealybug nymphs and lay 5 to 60 eggs inside each of the host body, depending on host size.

Life cycle

Fecundity	:	56-80
Total develop. period	:	14-15 days
Adult longevity	:	3-6 days
Total life cycle	:	14-16 days

They are highly sensitive to insecticidal sprays and hence needs a pesticide free environment for successful perpetuation.

Schedule and Method of release

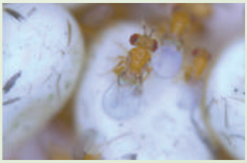
Inoculative release of the parasitoid @ 100 adults/acre as soon as the pest is noticed. Open the lid of the container and walk near the infested mulberry plants covering the whole garden so that the parasitoids escape and get access to the pest.

Trichogramma chilonis against Lepidopteran pests

There are about 150 species of *Trichogramma* worldwide that differ greatly in host preference, searching behaviour and tolerance to environmental conditions. They have been used in inundative releases more than any other natural enemy. They are tiny parasitoids that attack the eggs of over 200 species of moths and butterflies. When released in infested crop field, they seek out the eggs of the pest and lay their own eggs inside them. On hatching, the parasitoid larva feeds on the embryonic content of the host eggs and completes the development within. So, instead of a caterpillar, adults of *Trichogramma* will emerge from the pest eggs which again start searching for host eggs.

Once a female finds a host egg, it drills a hole through the chorion (egg shell) and inserts 2 to 3 eggs into the host egg. A single female can parasitize 1 to 10 host eggs/day or 10 to 190 during her life, thus providing excellent control of the pest. A parasitized caterpillar egg turns black in colour in 4-5 days after parasitisation as the young wasp develops inside it. Large females parasitize more eggs than smaller females.

Life cycle



Fecundity	:	35-100
Incubation period	:	1 day
Larval stage	:	3-4 days
Pupal period	:	4-4.5 days
Total develop. period	:	8-9 days
Adult longevity	:	Male 5 to 7 & female 4 to 12days

Schedule and Method of release

Release *Trichogramma chilonis* egg parasitoids @ 4 tricho-cards/ acre/crop (from July to February). Tear each card into small pieces and staple on the underside of mulberry leaves at random, covering the entire garden.

Nesolynx thymus against uzi fly

It is a hymenopteran gregarious ecto-pupal parasitoid. Life cycle is completed in about 15 days as follows:

Life cycle

Fecundity	:	250
Incubation period	:	1 day
Larval stage	:	7 days
Pupal period	:	7 days
Total develop. period	:	15 days
Adult longevity	:	6-8 days

Schedule and Method of release

2 pouches/100 dfls on 3rd or 4th day of 5th instar in rearing house. (From each pouch about 10,000 to 12,000 parasitoids emerge)

After the completion of rearing, they may be shifted to mounting/ harvesting place and finally to litter pit.

MASS PRODUCTION OF IMPORTANT BIO-CONTROL AGENTS

Mass production of ladybird beetles

Step-I: Production of mealybugs on sweet pumpkins

- Procure mature sweet pumpkins weighing about 2-3 kg having ridges, grooves & stalk and wash them with clean running water
- Surface sterilize by dipping in 0.5% Bavistin solution and air dry
- Seal/plug any damaged portion of pumpkin with melted paraffin wax
- Inoculate the sweet pumpkins with 50-75 gravid female mealy bugs or ovisacs so as to cover the entire upper half surface of the pumpkins
- Label the date of inoculation in each pumpkin
- Keep these infested pumpkins in a well-ventilated room on stainless steel stands and place them over iron racks
- Maintain the room temperature (25 to 28°C)
- In about 20 to 25 days, fully developed mealybug colony is obtained, which is ready for production of ladybird beetles

Step-II: Mass Production of predatory beetles

- In an aerated plastic container (18 cm dia.), release 10 pairs of one week old Cryptolaemus or 20 pairs of Scymnus beetles and provide 8-10 g of mealybugs. Also, provide a cotton swab dipped in 50% aqueous honey
- Adult beetles feed on the mealybugs as well as honey and start laying eggs in the mealybug colony

- In about 10 days, minute grubs of the beetles can be observed
- From 11th day to 22nd day, provide 3g mealybugs on alternate days
- From 23rd day, provide 1g of mealy bug on alternate days till majority of grubs settle for pupation
- After pupation, adults emerge in about 6 to 8 days
- Adult beetles continue to emerge for one week which can be collected by using aspirator and transferred to plastic containers
- Feed the fresh adult beetles with 50% aqueous honey in cotton swabs
- Place corrugated (folded) paper inside each plastic container for perching
- After 6 to 8 days, they may be released in the field or can be used for further multiplication.

It is estimated that from one pumpkin weighing 2 to 3 kg, about 75 to 90g mealybugs can be harvested from which about 125 *Cryptolaemus* or 250 *Scymnus* beetles (1 unit) can be produced. Hence, from 100 kg pumpkins, about 30 units of beetles are produced in 35-38 days.

Mass production of *Acerophagus papayae*

- Procure 2 months old potatoes and wash in clean water
- Disinfect using 5% Sodium hypochlorite solution
- Treat with 1 ppm Gibberlic acid solution (1 mg in 1 lit water) for ½ h
- Spread on a tray in single layer, air

dry, and keep for germination /sprouting in trays with sand layer

- Cover the tray with black cloth and keep for sprouting
- Within a week buds will sprout
- Infest the potato sprouts with nymphs of papaya mealybug
- Shift the infested potatoes to cages and release the parasitoid adults for parasitization. Cover with black cloth for incubation
- After about 10 days, transfer these potatoes with mummified papaya mealy bugs to wide mouthed plastic jars

- From 15th day onwards, parasitoid emergence starts in these jars which may be collected using an aspirator
- Provide honey (1:10) through plastic strips as food to these parasitoids
- Release them in papaya mealybug infested mulberry gardens as early as possible, as they are short lived.

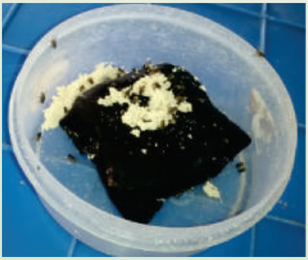
It is estimated that by utilizing 1 kg potato, about 20,000 parasitoids can be produced.

Mass production of *Nesolynx thymus* on housefly pupae

Step I: Production of housefly pupae

- Place 400 ml (about 14,000 pupa) housefly pupa in adult cage (an iron frame of size 45 cm x45 cm x45 cm covered with nylon net having 15-20 cm wide sleeve on one side)
- Approximately 80% flies will emerge in 4-5 days
- Maintain adult colony at 25±2°C temperature, 75±5% RH. and light duration 12:12
- Place 250 g of food containing 1 part sugar and 1 part milk powder
- Place cotton, soaked in water in the cage as a source of water, which should be ensured on daily basis


- Increase room temperature to 28±2°C and 90±5% RH, one hour prior to introducing the oviposition containers in 4-5 days old colony. (To prepare oviposition containers add 5-10g milk powder and 5g yeast powder to 100g of used larval medium and mix it thoroughly. Wrap in moistened black cloth and place in a plastic container. As an alternate, cotton soaked in milk and kept in plastic container can also be used for oviposition)




- After 5 to 6 h. remove the oviposition container which contains about 20,000 house fly eggs (2-3 ml)
- Gently remove the eggs from ovipostion container with a brush
- Place 5.5 kg of freshly prepared larval diet in plastic basin (45cm diameter and 15cm deep). To prepare larval diet, mix 1.2kg wheat bran and 15g dry yeast thoroughly. Add water to make final weight to about 5.5kg. Stir the mixture to ensure uniform distribution of water as well as all the ingredients
- Place 2-3 ml house fly eggs (about 20-25 thousand eggs) on the surface of larval diet and cover with kora cloth
- Eggs will hatch within 24h and the maggots start feeding
- After 5-6 days of feeding, maggots' colour changes into creamy white denoting initiation of pre-pupal stage




- Spread wire mesh (10 mesh size) on a perforated plastic tray (60cm x 30cm). Keep this tray on a non-perforated plastic tray (60cm x 30cm)
- Gently transfer the larval medium containing the pre-pupa on wire mesh and spread the medium
- All the pre-pupa will move down through wire mesh and fall into






the non-perforated tray kept below. Spread a thin layer of saw dust to facilitate pupation

- In 1-2 days, pre-pupa will change into brownish colored pupa
- Separate the pupa daily by sieving through wire-mesh (8 mesh size). The pre-pupa pass through the mesh and pupa remain on the surface of the wire mesh
- Determine the quantity of pupa by volume (one ml will contain about 30-35 pupae)

Step-II: Production of N. thymus on house fly pupae

- Spread 500ml (about 20,000 pupa) fresh house fly pupa in a single layer in 60 cm x 30 cm plastic tray

- Smear 50% honey solution on both sides of a plastic strip (20 cm x 2.5cm) and keep 2-3 such strips in each tray as food for parasitoid adults
- Release 4-5 thousand, one-day-old Nesolynx thymus adults in the tray in the ratio of 1:4 (1 female parasite: 4 house fly pupae). About 350ml parasitized housefly pupae will serve the purpose
- Cover the tray with thick white cloth and tightly fix it with elastic to prevent the escape of N. thymus
- After 4 days, collect the parasitized pupa from the tray and keep in adult cage for emergence of house fly, if any
- Remove the empty shells by winnowing and collect parasitized pupae for supply to stake holders
- 7-10 days old parasitized pupae may be mixed in equal quantities so as to achieve emergence continuously for a week

- After mixing, measure 50ml of house fly pupa parasitized by Nesolynx thymus and pack in 10cm x 10cm nylon net pouch
- From each pouch, about 10,000 adults of the parasitoid emerge. These pouches can be easily transported to stakeholders by courier
- Parasitized pupae (10-14 days old) can be refrigerated at 10±2°C for a maximum of one week in case of delay in supply, without affecting the quality of the parasitoids

Cost of establishment of mass production unit

Capacity/month: 2000 NT pouches & 200 units of beetles

Equipments	No.	Rate	Total Amount
Iron stand for basin: 6′x 3′ x 7.5′ (LBH) 9″ gap, 9 racks (1 rack holds 8 basins x 9 =72 basins)	1	8000	8000
Iron stands for plastic trays: 6′x2¼ x 7 (LBH) 6″ gap, 11 racks (1 rack holds 3 trays x 11 =33 trays)	1	6000	6000
Plastic crates (non perforated) 57 cm x 36 cm x 11 cm	20	500	10000
Plastic tray (non perforated) 90cm x 60cm x 6cm	30	500	15000
Plastic basins 45 cm diameter, 20 cm height	100	100	10000
Plastic tray (perforated bottom) (90 x 60 x 6 cm)	4	400	1600
Open shelf (6′ x2′ x 6′) for sweet pumpkin, jaly boxes & harvested beetles boxes (1′gap, 5 racks)	6	6000	36000
House fly cage 2′ x1.5′ x1.5′	20	300	6000
Plastic boxes with mesh lid (Jali box)	200 0	60	1200
Measuring cylinders: plastic (15 ml)	4	75	300
Measuring cylinders: plastic (500 ml)	2	300	600
Working table: wooden 2.5′ x 4′	2	1500	3000
Chairs: wooden/mild steel	4	750	3000
Stool: wooden/mild steel	4	400	1600
Wet & dry thermometer	2	1000	2000
Balance (ordinary)	1	1500	1500
Plastic container to store water	1	400	400
Stands for pumpkin	200	40	8000
Room heater (optional)	2	1500	3000
Refrigerator 250 lts (optional)	1	10000	10000
Office table with drawers-mild steel (optional)	1	3000	3000
Almirah (optional)	1	10000	9000
Total (Rs)			**1,50,000**

Consumables requirement per unit per month

Items	Beetles		NT	
	Qty	Amount (Rs)	Qty	Amount (Rs)
Sweet Pumpkin	700 kg	7,000		
Wheat Bran			500 kg	10,000
Milk powder			40 kg	10,000
Yeast			5 kg	1,150
Honey	1 kg	250	1 kg	250
Absorbent cotton	1 roll	150	2 rolls	300
Sugar			30 kg	1,200
Packing materials*		3,000		3,000
Cloth		200		1,000
Miscellaneous**		250		250
Total (Rs)		**10,850**		**27,150**
			Grand Total	**38,000**

* includes nylon net pouches, plastic boxes, cardboard boxes, gum tape *etc.*
** includes Soap, detergents, stapler pins, rubber bands *etc.*

Economics of production of bio-control agents

(Capacity/month: 2000 NT pouches & 200 units of beetles)

Monthly Expenditure	Amount (Rs.)	Sale proceeds	Amount (Rs.)
Establishment cost/ unit	1.5 lakh	2000 pouches of *N. thymus*@ Rs 50/Pouch	1,00,000
Raw material costs	38,000	200 Boxes of Cm/Sc beetles @ Rs 250/box	50,000
Building rent	4,000	Gross returns/month	1,50,000
Labour charges	12,000	Expenditure/month	54,000
Total	**54,000**	**Net profit/month**	**96,000**

Future of biological control

Chemical control cannot solve the problem of pests permanently, but biological control when works effectively, will solve the same permanently. Also, chemical control has several demerits. Hence, biological control has more importance in the management of pests. The international organizations like FAO, CIBC, USDA, IOBC, NBAIR etc, are coming forward to encourage the biological pest suppression. Therefore, biological control is in the lime light and widely accepted in the world and one can expect a bright future.

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