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INSECT PEST – RESISTANT VARIETY – NATURAL ENEMY: TRITROPHIC INTERACTIONS

SEMINAR REPORT

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ABSTRACT

Biological control agents and resistant cultivars are the major components of insect pest management programme. The physical, morphological and chemical factors of a resistant cultivar restrict the insect pest utilising the plant for normal feeding, oviposition and shelter. However, these factors can affect the effectiveness of the natural enemies also. In most integrated pest management programmes, the integration between these two strongly promoted tactics is not well known. In many instances, these tactics are complementary as well as supplementary. However, in some cases resistance mechanisms of the plant have been shown to hinder the effectiveness of natural enemies upon insect pests.

In cotton, frego bract cultivars are resistant to boll weevil, *Anthonomus grandis*. McGovern and Gross (1986) found that, the parasitism of boll weevil grub by larval parasitoid, *Bracon mellitor* was eight times more in frego bract cotton compared to the normal bract cultivars. In potato, the foliar pubescence, one of the resistant mechanism against potato tuber moth larvae, was known to adversely affect the parasitisation potential of the egg parasitoid, *Copidosoma koehleri* (Gooderham, 1999).

Giles *et al.*,(2002) found that, the predator *Coccinella septumpunctata* found in OKO8 cultivar of *Medicago sativa*, was having increased survival, development rate and bigger size compared to the ones found in cv. Windsor, due to difference in nutritional status. Effect of transgenic cultivars with *Bt* gene on natural enemies, was studied by several investigators. Zwahlen *et al.*,(2000) found that there was no adverse effect of transgenic *Bt* corn on the natural enemies *Orius majaculatus* and *Chrysoperala carnea*. Hence, a through understanding of tritrophic interaction is inevitable for a successful pest management programme.

INSECT PEST – RESISTANT VARIETY – NATURAL ENEMY: TRITROPHIC INTERACTIONS

INTRODUCTION

Biological control agents and resistant cultivars are major components of insect pest management programme. Breeding of resistant cultivars include introduction of several defensive traits into the cultivated species, which are disadvantageous to the herbivore. The physical, morphological and chemical factors of a resistant cultivar restrict the herbivore utilising the plant for normal feeding, oviposition and shelter. But these factors can affect the effectiveness of the natural enemies also. The compatibility of biological control and plant resistance has been examined for several plant - herbivore - natural enemy complexes. (Price *et al.*, 1980). In most integrated pest management programmes, however, the integration between these two strongly promoted tactics is not well known. In many instances, these tactics are complementary; for example, plant resistance may slow the rate of increase of a pest population, thus allowing biological control agents to provide effective control (Mc Auslane *et al.*, 1995). In contrast, many resistance mechanisms have been shown to hinder the effectiveness of natural enemies (Ramnath *et al.*, 1992). In this context, this presentation will focus on the scientific and pragmatic issues related with the integration between resistant traits and natural enemies, and combining the both for a successful pest management programme.

PLANT CHARACTERISTICS THAT AFFECT NATURAL ENEMIES

Plant characteristics can influence the predators and parasitoids of insect pests in two primary ways.

1. Direct plant effects on natural enemies.
2. Indirect plant effects mediated through the prey.

Direct plant effects entail only the interaction between the plants and natural enemies. These effects involve simple mechanisms. Whereas, indirect plant effects result from interactions between the plant, herbivores, and natural enemies. The indirect effects often involve complex interactions and may not be well understood.

Bottrel and Barbosa (1998) have outlined the various plant characteristics that affect the natural enemies.

A. DIRECT PLANT EFFECTS

1. Plant morphology:
   - Plant size and plant part shape
   - Epicuticular waxes
   - Pubescence
2. Plant semiochemicals:
   - Attractants
   - Repellents
   - Inhibitors
   - Toxins
3. Plant Nutrition

B. INDIRECT PLANT EFFECTS

1. Semiochemical mediated interactions
2. Herbivore sequestration of plant allelochemicals

A. DIRECT PLANT EFFECTS

1. Plant morphology

Several investigators have examined the influence of plant morphology on entomophagous spices. Doggett (1964) concluded that, closed panicles of certain sorghum varieties protected the *Helicoverpa armigera* from larval predators and parasitoids. Similarly, the type of leaf configuration, open vs. closed, in cabbage cultivars was observed to affect host finding and parasitism of *Pieris rapae* by *Phryxe vulgaris* and *Apanteles glomeratus* (Pimental, 1961). Further, Berlinger and Golberg (1978) reported that, the shape of fruit sepals had a direct effect on the parasitism of the citrus mealy bug, *Planococcus citri* by the encyrtid, *Anagyrus pseudococci*.

In cotton, bract shape and size influences the ability of parasitoids and predators. Mc Govern & Gross (1976) reported that parasitism of boll weevil grub, *Anthonomus grandis* by *Bracon mellitor* was eight times greater in frago bract (58%) compared with normal bract (7%) in cotton. (the former, a cultivar in which the bracts are twisted and do not encase the bud). As a counter example, Mc Govern & Gross (1976) also found that parasitism of boll weevil grub by a second species, *Hetrolaccus* *grandis* was greater in normal bract (14%) than frago bract (8%) cotton. Regarding the ability of predators in the above cultivars, Lincoln *et al.* (1971) found that mortality of *H. Zea* due to predation averaged 95% on frago bract compared with 50% on normal bract cotton. So, one need to be careful in generalising across all predators and parasitoids in relation to a cultivar.

Surface Wax

Surface waxes also affect the searching efficiency of several predators. Carter *et al.* (1984) found that, the larvae of coccinellid, *Coccinella septempunctata* dropped more often from smoother pea leaves, compared to cultivars having rough leaf surface. The tarsal setae of coccinellids apparently stick more readily on smooth surfaces than on rough surfaces.

Trichomes

Much widely studied interaction is between the trichomes (hair like appendages extending from the epidermis of aerial tissues of plants) and entomophagous species. These trichomes may be of uni or multi cellular, glandular or non-glandular, and of several morphological types such as straight, hooked or stellate. Foliar pubescence occurs on a wide variety of agricultural crops and its role as plant defensive mechanism has been demonstrated for several plants

Influence of non-glandular tricmomes

In a detailed study of the searching behaviour of the greenhouse whitefly parasitoid, *Encarsia formosa*, on pubescent and non pubescent cucumber varieties, Li Zhao Hua *et al.* (1987) in Netherlands demonstrated that, the walking speed of this parasitoid was inversely related to trichome density. On cucumber (cv 'I.V.T no 71240'), numerous hairs on the leaf surface hampered the tiny wasp and reduced its walking speed, which makes it more time consuming to find hosts. The large hairs also trap large amounts of honeydew deposited by whitefly on leaves. Wasps that have run into a droplet of honeydew spend much time in preening their bodies. These observations suggested that on cucumber, biological control of the whitefly with *E. formosa* could be improved by reduction of the hairiness. A hairless mutant (I.V.T. no 761077 - Mayak) was introduced in the Netherlands by De Ponti (1988). It appeared however, that *E. formosa* were walking so fast on the hairless varieties, that they merely walked over whitefly larvae without noticing them. Therefore research was continued with experimental hybrids having a hair density of 50% of that of current commercial cultivars. These hybrids can be easily developed, as the inheritance of glabrousness appeared to be intermediate.

S.Ramnath and S.Uthamasamy (1992) studied the interactions of host plant resistance and natural enemies for the management of the *Helicoverpa armigera* on cotton. They had taken six cotton genotypes -MCU 11, MCU 9, TCH 1002, LPS 141, LK 861 and JK 276-4 that differed in hairiness of leaf. The efficiency of two natural enemies, *Chrysopa scelestes* (Banks) and *Trichogramrna chilonis* Ishii was tested against the egg and neonate larvae of *H. armigera* in lab condition.

Table 1. Integration of cotton genotypes and natural enemies on eggs and larvae of *H. armigera*

Genotype	Trichome density (No./cm^2)	Predation (Egg/day)	Parasitisation (%)
MCU 9	92	9.33	44.20
MCU11	33	17.33	60.00
TCH 1002	107	9.00	33.33
LPS 141	16	17.33	65.00
LK 861	10	18.67	74.20
JK 276	126	7.67	30.00

As regards the predation of eggs and neonates by *Chryropa scelestes*, genotypes with fewer trichomes recorded increased percentage of predation. Egg predation was maximum on LK 861, which had minimum number of trichomes on the leaves. The findings are in conformity with that of Treacy *et al.*, (1983) who found that predation of *H. zea* eggs by *Chrysopa rufilabris* was low in pilose varieties and high in glabrous cotton. Cotton leaf trichome inhibited the movement of the predator grub over leaf surfaces and reduce the predating ability of the aphidline grub. Treacy (1987) also reported that the third instars were less affected by trichomes than were first and second instars.

Parasitisation by *T. chilonis* was also maximum on genotype with fewer number of trichomes. The parasite being a small insect gets entangled with in trichomes and reduces the ultimate parasitisation level. Similar adverse effect of foliar pubescence on the egg parasitoid *Copidosoma koebleri* on potato tubermoth was reported by Gooderham (1998). The parasitoid located the greatest number of host eggs *(Pthorimaea opercullela)* on the cultivars with least dense trichomes.

On the other hand, trichomes may favour some natural enemies. For example, dense hairs on leaf axils (domatia) provide hiding places for some predatory mites. In grapes, spidermites (Tetranychidae) are the major pest world over. But the predatory phytoseiid mites often prevented outbreak of these spidermites. Duso (1993) demonstrated that two important phytoseiid predators, *Typhlodromus pyri* and *Amblyseius aberrans* respond to leaf surface structure. A comparison of grape varieties having either glabrous or densely hairy leaf under surface showed that, both predators were consistently more abundant in hairy varieties. This difference was especially evident under hot, dry season when grapevine with hairy leaves acted as humid refugia for a large population of predatory mites.

Skirin and Fenlon (2001) found that the predatory mite *Phytoseiulus persimils* was more active on smooth leaves of *Choisya ternata* and efficiently checked the population of *Tetranychus urticae*

Influence of glandular trichomes

Glandular trichomes secrete sticky substances that may inhibit movement or produce chemicals that are repellent or toxic to natural enemies. Glandular pubescence, a non specific arthropod resistance mechanism in the wild potato, *Solanum berthaultii*, is being used in a potato breeding programme as a defence against a variety of insect pests, including *Myzus persicae*, *Empoasca fabae* and *Leptinotarsa decemlineata*. Highly resistant potato hybrids *(S. tuberosum* x *S. bertbaultii)* have been produced from this breeding programme, which bears high levels of glandular trichomes.

Obrycki (1986) conducted laboratory, green house and field studies to assess the interaction between the potato hybrids bearing various densities of glandular trichomes and natural enemies. Under green house conditions, there was a direct relationship between the density of glandular trichomes and efficiency of predators. The mobility of newly hatched coccinellid and chrysopid larvae was inversely related to the density of glandular trichomes. On *S. tuberosum*, larvae moved over 100 mm in 48 hours, while on the pubescent *S. berthaultii*, movement was reduced to less than 10 mm.

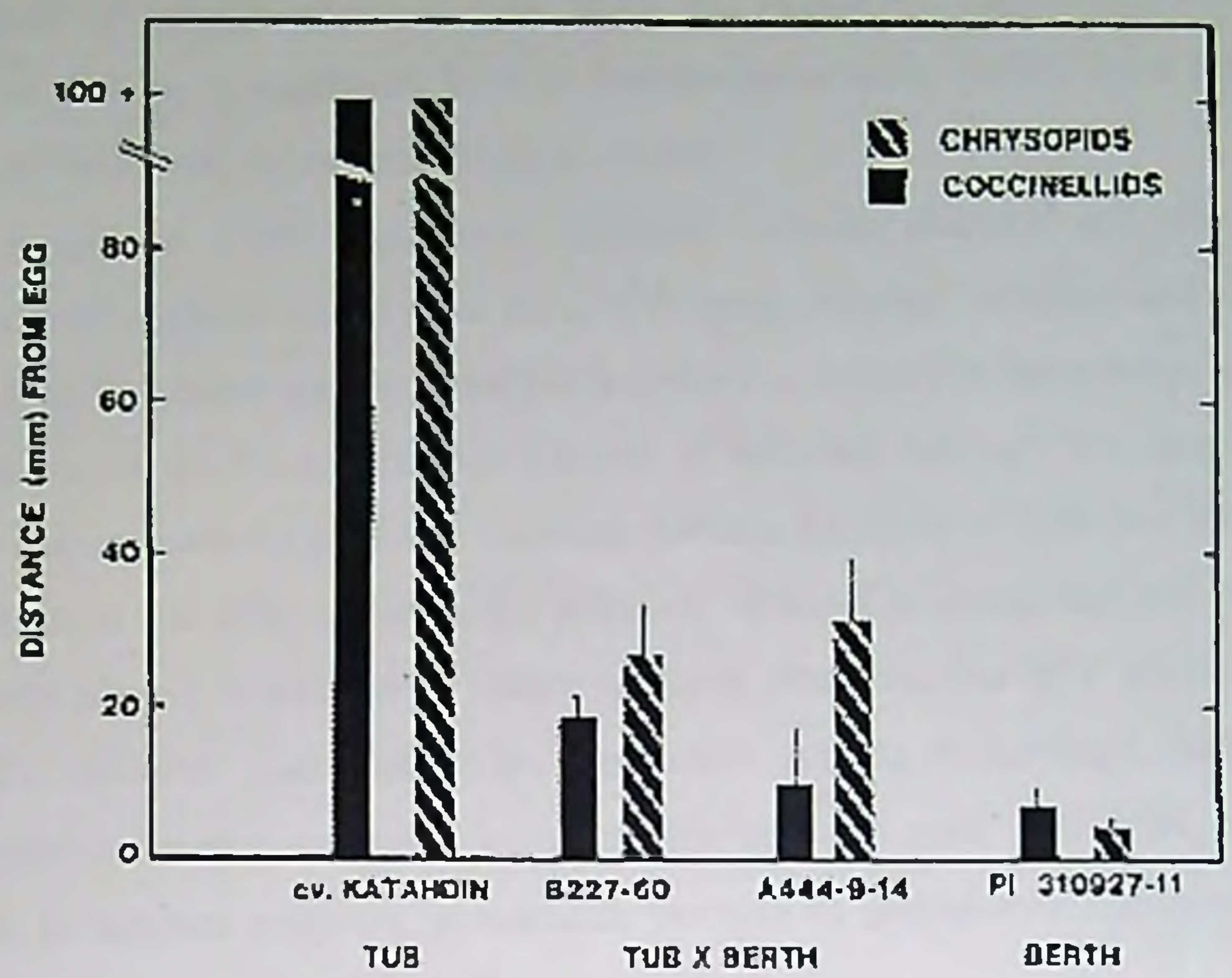

Fig.1. Movement of *Chrysopa* sp. and *Coccinella septumpunctata* on potato cultivars with various trichome densities (Obrycki, 1986)

However, these severe negative effects observed under greenhouse condition were attenuated in the field. The same coccinellid and chrysopid species that were virtually excluded in the greenhouse studies were present and oviposited on the pubescent clones in the field conditions. This may result from the effects of dust, wind and rain on the glandular trichomes, and the detrimental effect of these trichomes was considerably reduced in the field conditions.

2. Plant Semiochemicals

The olfactory system of a parasitoid or predator must accomplish several tasks before the natural enemy can find and attack its host or prey. Plant volatiles may be especially important in guiding parasitoids and predators to their prey habitats. Many parasitic insects and predators can distinguish the volatiles of closely related plant genotypes in laboratory experiments.

Genotypes of a crop species may differ significantly in the production of specific volatiles. Glanded cotton *(Gossipium hirsutum)*, for example produces about 100 times more of the synamones, that attracts the parasitoid *Campoletis sonorensis* than the non-glanded cotton (Elzen *et al.*, 1985). Lewis *et al.*, (1985), emphasised the importance of several chemical cues from the plants and herbivores in increasing parasitoid activity. Compounds such as hexadecanoic acid, mostly from plant origins improved the activity of entamophagous species.

Annadurai (1992) extracted different volatile fraction and allelochemical complex from various cotton plant parts in different resistant cultivars and assessed the response of *Chrysopa scelestes* and *Trichogramma chilonis* in laboratory bioassay and in green house condition. Volatile fraction of different cultivars was extracted using hexane and assessed by petridish bioassay. Hexane fractions of different cultivars were sprayed on to the filter paper in the petridish, allowed to evaporate and *Helicoverpa* eggs were placed in petridishes before bioassay. Five females of *T. chilonis* and five grub of *C. scelestes* were used for the experiment. Among the cultivars, both parasitism and predation rates were higher in susceptible cultivars such as SUVIN, while it was reduced in resistant cultivars, presumably because of undesirable chemical factors, as evident from chemical profiles of various cultivars.

Table 2. Percentage predation and parasitism on *H. armigera* in cotton cultivars.

Cultivar	Predation by *Chrysopa*	Parasitism by *Trichogramma*
Suvin	64.9	52.7
LRA 5166	52.6	45.8
MCU 11	50.4	45.4
TCHB 213	38.0	39.4
MCU 7	31.7	36.5

Rapusas *et al.* (1996) tested the intraspecific variation in chemical attraction of rice to insect predators *Cyrtorhinus lividipennis* (Miridae) and *Micraspis hirashimai* (Coccinellidae). They tested 15 rice cultivars showing variable resistance to the brown planthopper, *Nilaparvatha lugens*. The predators were attracted towards the volatiles from susceptible cultivar TN1, and the resistant variety IR 72 showed less attraction.

Havill and Raffa (2000) evaluated the activity of female parasitoid *Glyptapantelus* on *Populus* trees. They found that, the parasitoids were attracted towards the pest *Lymantria dispar* damaged leaves compared to undamaged leaves.

3. Plant Nutritional Effect

Many natural enemies feed directly on plants or plant products such as pollen, floral nectar, and extra floral nectar. Only about 25 per cent of the 163 families of parasitic and predatory insects, that Hagen (1987) examined were strictly carnivorous. The remaining 75 per cent contained species that may consume plant materials during the life. The plant materials allow natural enemies to survive periods of host or prey scarcity and contribute to a nutritionally balanced diet (Coll, 1996). Some predatory ladybird beetles can complete their life cycle when pollen as the sole food source (Shepard, 1991). Pollen can sustain the predatory anthocorid, *Orius insidiosus* for long periods. *O. insidiosus* and the predatory coccinellid, *Coleomegillia maculata* reach highest densities in corn during pollination. The availability of corn pollen, along with alternative prey (aphids, thrips) increased their success in controlling the European corn borer, *Ostrinia nubilais* (Coll, 1992).

Cotton has been the main crop plant, investigated for the role that extra floral nectar play in supporting both herbivorous, entomophagous insects and spider mites. The presence or absence of extra floral nectar can influence the diversity and abundance of entomophagous species occurring in cotton fields. Lack of this extra floral nectar on cotton is a highly desirable trait, because it suppresses important lepidopterous and hemipterous pests. But the total population of natural enemies were reduced from 17 to 35 per cent in fields of extrafloral nectariless cotton (Schuster *et al.*, 1976). Significant reductions were found in numbers of *Chrysopa carnea*, *Orius insidiosus*, and *Coleomegilla maculata* in these cultivars.

De Lima and Leigh (1984) evaluated the relationship of nectar to the predator, *Geocoris pallens* and *G. puncticeps* respectively. Development of *G. pallens* at all stages was reduced greatly when the insects were deprived of both prey and nectar. Longevity was increased considerately when the insects were provided with nectar

Table 3. Longevity in days of *Geocoris pallens* nymphs and adults deprived of prey in different cotton genotypes.

Stage	Acala SJ2 (Nectaried)	H 6124 (Nectariless)
2nd instar	30.4	7.9
3rd instar	31.3	8.0
Adult	52.4	16.0

B. INDIRECT PLANT EFFECTS

Indirect plant effects results from the indirect interactions between the plants, herbivores and natural enemies. Because all the arthropod pests of crops derive their nutrition from plants, the natural enemies using the pests as prey indirectly get their nutrition from plants. The food source will affect the size, survivorship, development time and density of the herbivore; in turn these factors may affect natural enemy size, survivorship, development time and density also. This tripophic relation often involves complex interaction and may not be well understood.

Morgan (1910) was the first to consider, the natural products in foliage may affect the success of entomophagous parasitoids. He proposed that, the reduced percentage parasitism of *Manduca sexta* larvae on tobacco plants by the parasitoid *Apanteles* sp. resulted from the ingestion of nicotine by the host and subsequent toxification of the parasitoid. Following up the studies of Gilmore (1938) showed that when larvae of *M. sexta*, fed upon a variety of tobacco foliages with differing level of nicotine, the hosts were parasitised at different levels. Thus, the plant defences against the herbivores such as poor nutritive value, low soluble nitrogen and intrinsic toxins are sequestered into the natural enemy through the herbivore and affects the physiology of natural enemy also.

Heneidy *et al.* (1988) studied the influence of dietary nicotine on the fall armyworm, *Spodoptera frugiperda* and its parasitoid *Hyposoter annulipes*. Early workers have demonstrated the effects of a single concentration of nicotine in host diet on two parasitoid species, *Cotesia congregata* and *Hyposoter annulipes* (Barbosa *et al.* 1986). However, no data are available on the trends in development and survival of parasitoids, exposed to a range of concentration of dietary nicotine or on the influence of these concentrations across generations of the parasitoid. Heneidy *et al.* (1988) conducted a laboratory experiment with differing levels of nicotine. Neonate larvae of *S. frugiperda* were placed on the semi synthetic diets containing the following concentrations of nicotine 0.0%, 0.025%, 0.050% and 0.075%. Three to four day larvae were parasitised using *H. annulipes* and the development and mortality of the parasitoid were observed. In each nicotine concentration, emerging parasitoids were used to parasitise fall armyworm larvae reared at the same concentration, thus producing another generation of parasitoids.

The results revealed that, the greater the concentration of nicotine in the diet of fall armyworm hosts, the greater the mortality of *H. annulipes* larvae. Larval and total mortality were also greater in later generations. Parasitoid size was also adversely affected by nicotine. As dietary nicotine increased, the dry weight of both male and female parasitoids dropped significantly. Parasitoids of the second and third generations had greater mortality, longer development and smaller size compared to the fist generation. Reduced prey quality does not necessarily diminish the effectiveness of natural enemies. For example, soybean cultivars resistant to the Maxican bean beetle *Epilachna varivestis*, increased the development time and reduced the reproduction of the beetles parasitoid *Pediobius foveolatus* (Kauffman, 1985). However, these negative effects did not diminish the parasitoids potential for biological control, because its reduction in population growth potential was less than that of its beetle host.

Giles *et al*., (2002) studied the interaction of two *Medicago sativa* cultivars, aphid prey *Acyrthosiphum* and the predator *Coccinella septumpunctata*. Compared with the aphid predator reared on Windstor variety of *Medicago*, the aphids reared on cv. OKO8 stored significantly stored more fatty acids (1.17 fold more). This in turn increased the survival, development and size of the predator *Coccinella septumpunctata* on OKO8 cultivar.

MULTIPLE EFFECTS OF PLANTS

Plants may have multiple effects on natural enemies. For example, rice cultivars resistant to the brown plant hopper, *Nilaparvatha lugens* reduced the insects growth and stimulated increased probing and movement on plants. Sengutluvan and Gopaan (1990) reported that, the mirid predator *Cyrtorhinus lividipennis* was more active against the BPH in resistant rice cultivars, which they attributed to the greater plant hopper activity on resistant cultivars.

Reitz and Trumble (1999) evaluated the tritrophic interaction among *Apium* sp., herbivore *Trichoplusia ni* and parasitoid *Copidosoma*. Secondary metabolites, furanocoumarin present in the plant is a resistant factor for *T. ni*. When the *Copidosoma* was allowed to parasitise the furanocoumarin fed *T. ni*, a increase in mortality of the parasitoid was noticed.

Haseena (1999) studied the population dynamics of natural enemies in three different rice cultivars namely, Jaya, a susceptible variety; Jyothy, a moderately resistant variety; Kanakom, a resistant variety. The populations of predators

Cyrtorhinus lividipennis and *Micraspis* sp. were found to feed more BPH when reared on resistant variety. Similarly, the activity of predatory spider, *Lycosa psudoannulata* and *Microvelia* sp. were more on resistant varieties.

EFFECTS OF TRANSGENIC CULTIVARS ON NATURAL ENEMIES

The first genetically engineered corn, cotton and potato cultivars containing the gene from the bacteria *Bacillus thuringiensis* var. *kurstaki*, that encodes for the expression of insecticidal endotoxin were commercially produced in the United States. The safety of these transgenic plants to the non-target organism is currently of great concern. Pilcher *et al.* (1997) studied the development, survival and field abundance of three predators, *Coleomegilla maculata, Orius insidiosus* and *Chrysoperla carnea* on the transgenic corn cultivar. They observed no adverse effect on the three predators by the transgenic corn cultivar with *Bt* gene .

Johnson and Gould (1992) examined parasitism of *Heliothis virescence* on transgenic tobacco and reported that transgenic tobacco appeared to be compatible with two larval parasitoids, *Compoletis sonorensis* and *Cardiochiles nigriceps*. Sims (1995) reported no adverse effect of transgenic *Bt* cotton *Gossypium hirsutum* [Cry I A(c)] on several beneficial insects, including *Apis mellifera, Nasonia vitiripennis* and C. *carnea*. Zwahlen (2000) found *Bt* corn was found to be safe for the parasitoid *Orius majaculatus*.

CONCLUSION

Both biological control and plant resistance constitute two of the most important integrated pest management tactics utilised today. It is extremely important that, the significance and complexity of interactions between these two traits should be evaluated thoroughly before introducing a resistant variety (Obrycki, 1986). Although plant breeding programme world wide are incorporating pest resistance traits into crops, they are doing so without considering, how pest resistance or other plant traits may influences natural enemies.

A better understanding of plant-pest-natural enemy evolution is necessary to predict, how to combine natural enemies and plant resistance for a long term results (Bottrell, 1998). Another need is the determination that, how much a natural enemy enhancing trait contributes to increased biological control in field condition. Painter (1951) stated that, any relationship between resistant plant varieties and insect parasitoids cannot be predicted with certainty beforehand but will need to be worked out in each individual case. Working out these relationships promote the use of these two tactics in a compatible and complementary manner in the integrated pest management programme.

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DISCUSSION

Q1. What are semiochemicals?

Chemicals involved in the communication among organisms are known as semiochemicals. For example pheromone is a semiochemical which is used for the intraspecific communication and allomone is another semiochemical used for interspecific communication.

Q2. Is it possible to include one more trophic level to tritrophic interaction?

We can include hyperparasitoids, that is paasitoid of a parasitoid, as the fourth trophic level to an existing tritrophic interaction. But it will be more complex to study the interactions involving four levels. This can be done for key pests of economically important crops.

Q3. Whether tritrophic interaction indicates only the interaction among insect pest, resistant variety and natural enemy?

Tritrophic interaction is a common term indicating the food chain base interactions among any three trophic levels. In this presentation, the interaction among insect pest, resistant variety and natural enemy is taken in to account.

Q4. Is there any relation between tritrophic interaction and environmental factors?

Study of tritrophic interaction involves only food chain based relationship and other factors are not taken into account.

POPULATION DYNAMICS OF TEA MOSQUITO BUG AS INFLUENCED BY WEATHER

By

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(2001-11-41)

SEMINAR REPORT

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ABSTRACT

Cashew cultivation in our country is given a prime importance from the last one-and-a half decades due to its high export value. Recently cashew production in our country is not in tune with increase in area under cultivation. This is due to the incidence of tea mosquito bug which is a serious pest of cashew across the west coast, while it is totally nil in the north-eastern region. This emphasise the influence of weather on population build up of tea mosquito bug.

The most favourable period for rapid multiplication and population build up of the pest is in between December and February synchronising with flushing and flowering in cashew. The population of tea mosquito is low in April and May and the pest is totally absent during rainy season when the crop is in its dormant stage. The population build up of the pest was found to be negatively correlated with the meteorological factors like minimum temperature, minimum relative humidity and rainfall and positively with sunshine.

If sufficient data on pest surveillance, phenology of the crop under varied environmental conditions, seasonal biology of the pest under different field conditions etc. are available, pest forecasting models can be made which helps in efficient pest forewarning. If we can predict the pest outbreak in advance it will be a boon to the farming community. This approach emphasizes need based sprays and reduces environmental problems to a greater extend.

POPULATION DYNAMICS OF TEA MOSQUITO BUG AS INFLUENCED BY WEATHER

INTRODUCTION

Cashew, the poor man's crop and the rich man's food is an important cash crop in India. It is also known as a coastal tree since the cultivation is mainly confined to the west and east coasts of India. Cashew cultivation in our country is given prime importance from the last one-and-half decades due to its export value. Recently cashew production in the country is not in tune with increase in area under cultivation. There was a drastic decline in cashew production even though the area under cultivation is increased to a greater extent. Decline in production is mainly attributed to the incidence of tea mosquito bug *Helopeltis antonii* Signoret, which is the most serious pest of cashew across west coast causing inflorescence blight and drying up of tender shoots and nuts. TMB alone can cause a loss upto 50% or more. The incidence of this pest is highly severe in the west coast of India, while it is totally nil in the north-eastern region. From this it is very clear that weather plays an important role in the population build up of TMB.

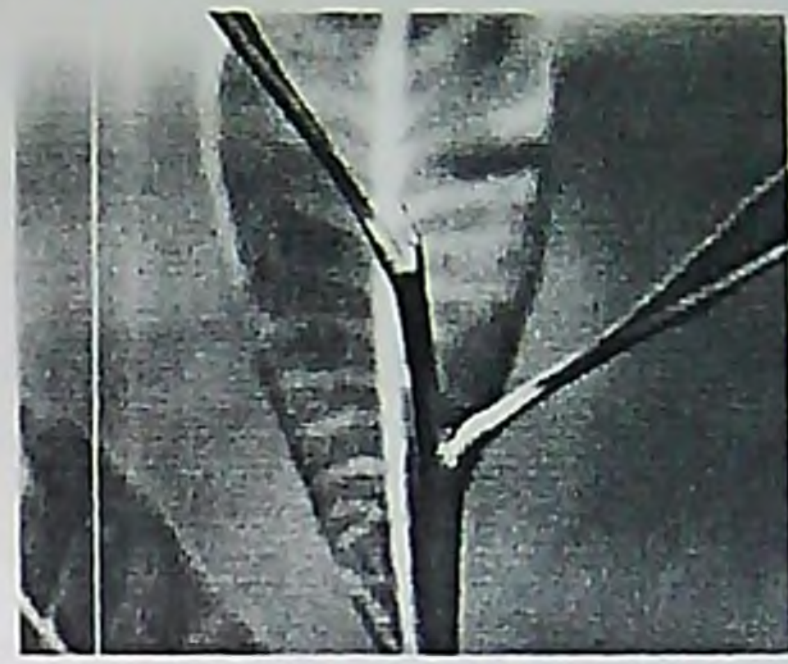
Then TMB incidence is expected to be severe in most of the cashew growing tracts of Kerala, Karnataka, Goa and Maharashtra on the west coast and also in Guddalore, Vridhachalam and Pudukottai areas of TN in the East coast. In Kerala, a bumper crop of two lakh tonnes of cashew nut was expected in 1998-99 as profused flowering was noticed in Dec-Jan due to favourable weather. However, the crop was severely damaged due to the incidence of TMB and the crop harvested was only half of the expected (Prasadarao, 2002). The insect also occurs on guava, cocoa, neem etc. (Puttarudriah and Appanna, 1955) and it can therefore multiply uninterruptedly throughout the year.

Nature and Symptoms of damage

Both adults and nymphs suck sap from tender shoots, panicles and immature fruits. The typical feeding damage is the formation of a necrotic area called

SYMPTOMS OF DAMAGE

On seedlings



On tender shoots

On panicles

On tender nuts and apples

On adult tree

lesion around the point of entry of the stylets of the mouth parts of the bug. The lesions appear as water soaked areas soon after feeding and became prominent, turning brownish black due to the death of plant cells. The lesions are elongate on tender shoots, its size varying with the maturity of the affected tissues. When the shoots are tender, the adjacent lesions coalesce and the entire shoot dries up. On tender leaves the pest infestation results in the crinkling and malformation of leaves Feeding marks are seen all over the leaf lamina in the form of brownish black lesions. In severe cases of infestation especially during the emergence of new flushes the infested trees present a scorched appearance. When the panicles are tender, the lesions coalesce and the entire panicle dry up. Very often the affected shoots and panicles are associated with the fungus *Colletotrichum* sp resulting in shoot die back and drying up of inflorescence. The damage on the fruits occurs on immature nuts and apples. The lesions on the fruits appear as brownish-black circular scaby spots. When immature nuts are infested, they shrivel and dry up. When the infestation occurs during the early stages of fruit set, it results in immature fruit drop

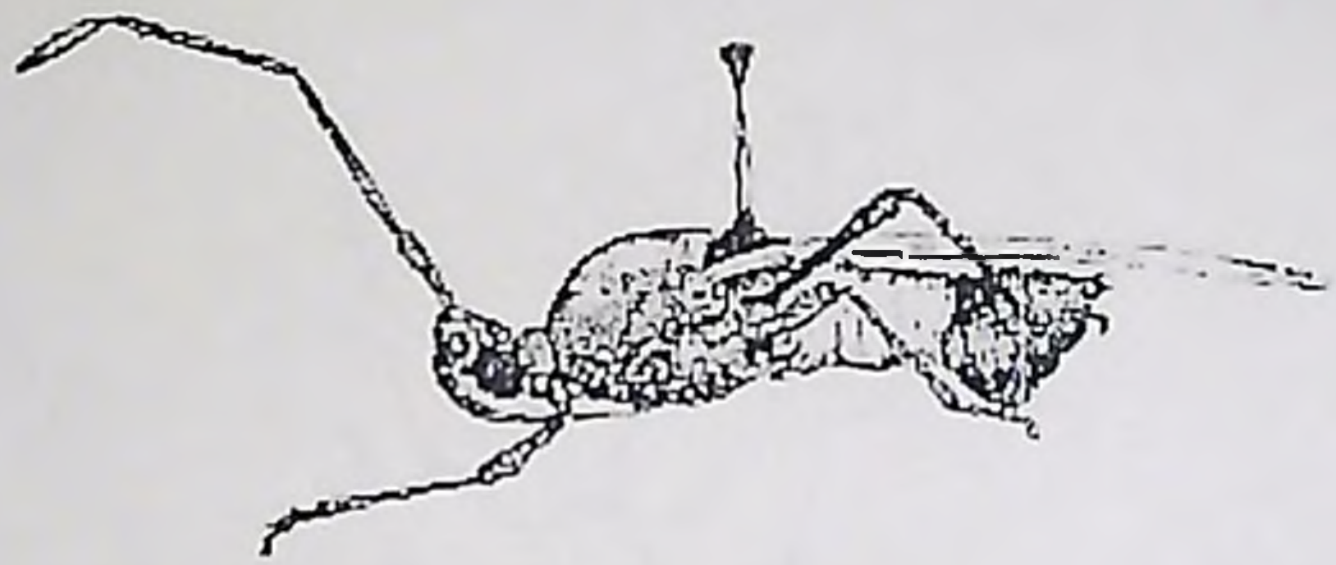
The feeding activity of the pest is higher during the morning hours before 9 am and in the evening hours after 4 pm (Abraham and Nair, 1981). Final instars nymphs (both 4th and 5th instar nymphs) can be more damaging than the adults. A final instar nymph was found to cause 114 lesions (range 78-235), an adult female 97 lesions (range 16-238) and the adult male 25 lesions (range 11-59) during 24 hour period.

Morphology

The morphometrics of adults and immature stages of *H. antonii* have been presented by Ambika and Abraham (1979). Adult female measures about 8 mm in length and 0.76 mm across the thorax, the adult male is slightly smaller, measuring 6 mm in length and 0.74 mm across the thorax. A knobbed process arises from the dorsal aspect of the thorax of both sexes, which is erect, tapering, apex being swollen and funnel shaped.

Male

Female

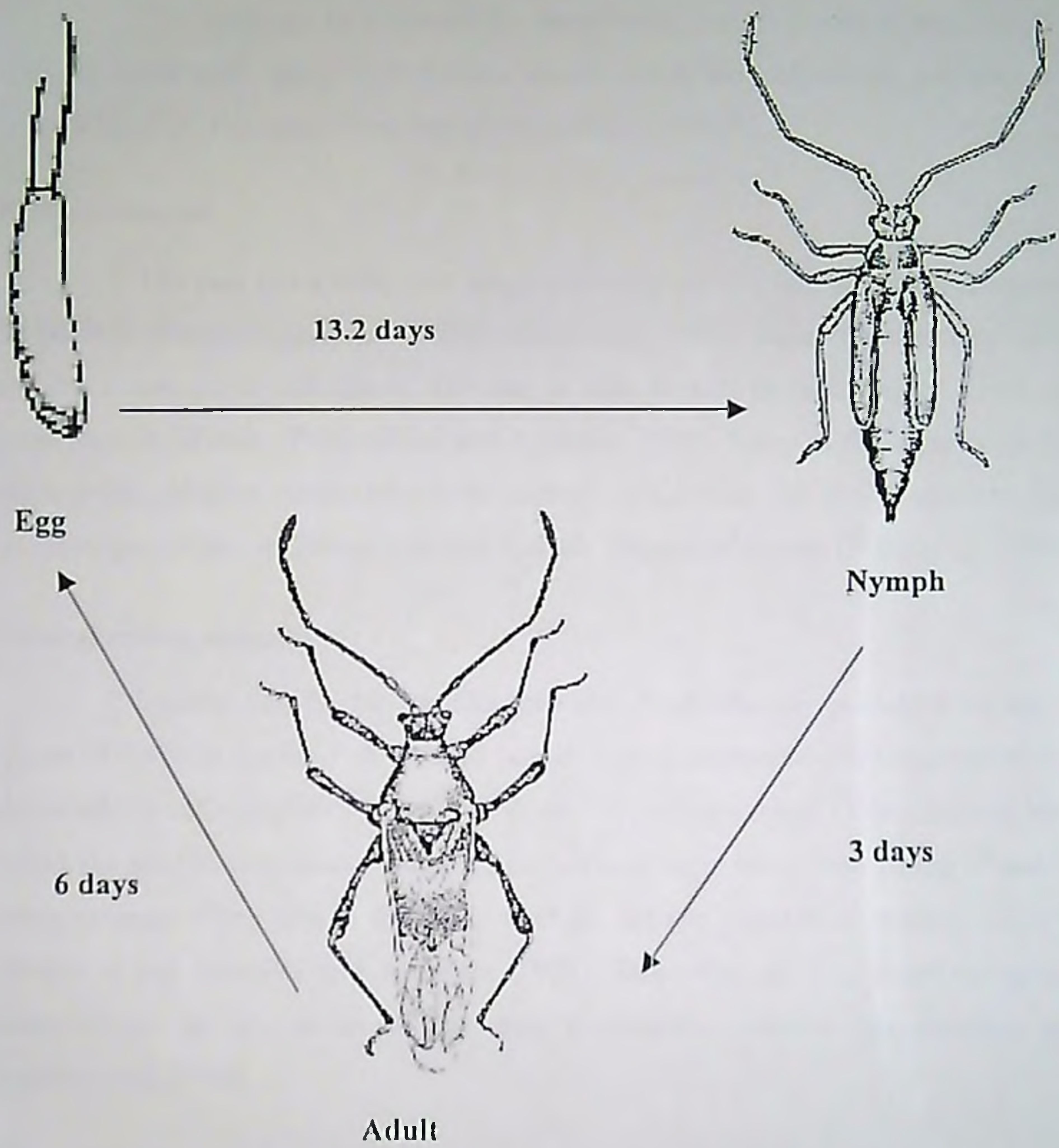


Lateral view

Biology

The biology of *Helopeltis antonii* was studied under lab conditions (Ambika and Abraham, 1979). The mean longevity of female is 6.5 days while the male life span lasts for 5.2 days only. The fecundity of female is 13-82 eggs and was maximum during the period Jan-Mar. Incubation period of eggs ranged between 6-10 days and was maximum during July-Sept.

Eggs are laid inserted into tender shoots and inflorescence stalks and sometimes on the leaf mid rib and petioles either singly or in groups of 2-6. The site of oviposition can be recognized by the presence of a pair of silvery white thread like chorionic process of unequal size that projects out side the plant tissue. There are five nymphal instars. Nymphal period lasts for 10 days (at a temperature range of 24-32^{0}C and relative humidity of 47-100%)



Periodical sampling of field populations revealed that in nature female always predominate, the range in the ratio of female to male being from 1:0.49 to 1:0.62. The pre-oviposition and oviposition periods at 25$\pm$0.5^{o}C lasted for 4 and 6 days respectively. The life cycle from egg to adult emergence occupied 22.3 days at

$28\pm1^{\circ}C$, the duration of different stages being 1.3, 2.1, 3.5, 3.2, 3.3 and 2.8 days for egg and nymphal instars I, II, III, IV and V respectively (Ambika and Abraham, 1979)

The optimum temperature for fertilization and oviposition was $25\pm0.5^{\circ}C$ while for embryonic and post embryonic development, the temperature preferred was around $28\pm5^{\circ}C$. The oviposition was suppressed at $31\pm0.5^{\circ}C$.

Host association

The pest has a wide host range occurring on 17 plant species representing 13 families (Devasahayam and Radhakrishnannair, 1986). Most common host plants are tea, cocoa, neem and guava. The pest is also seen to be attacking on apple and grapevines in Mysore (Puttarudriah and Appanna, 1955). Some of the spice crops like black pepper,allspice etc are seen to be harbouring the pest. The pest is also observed on *Moringa oleifera* in Trivandrum and Kollam districts of Kerala (Pillai *et al*., 1979)

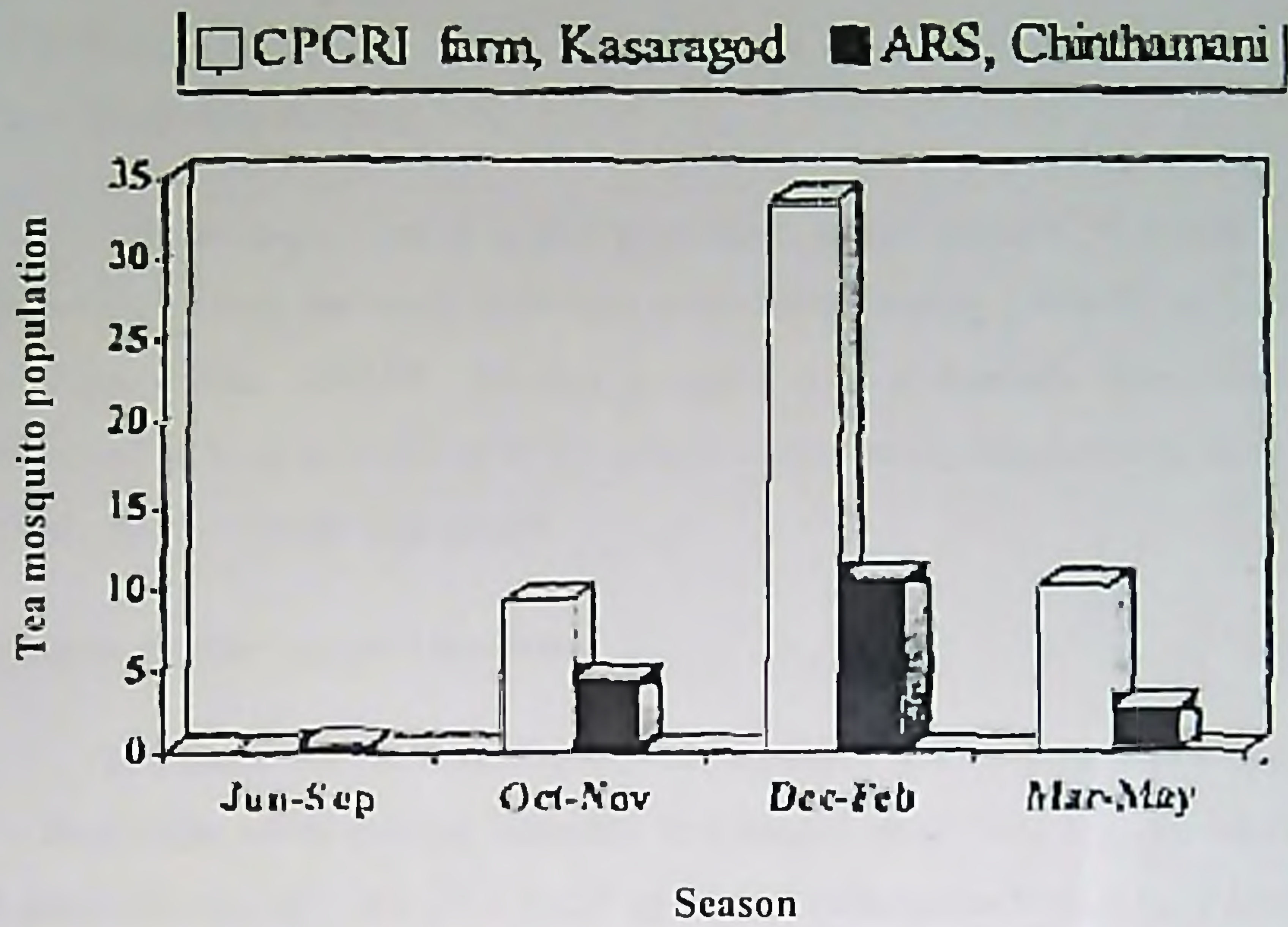
Natural enemy complex

Spiders like *Hyllus* sp, *Oxyopus* and *Phidippus* are predatory on earlier instars of TMB in the field. A reduvid bug *Endochus inornatus* was observed to feed on the adults and nymphs of the pest at Vittal (Devasahayam and Radhakrishnan Nair, 1986). An ant *Cremastogaster wroughtonii* has been recorded to feed on the 1st and 2nd instar nymphs of the pest in the field. A single ant was capable of feeding about 10 nymphs a day (Ambika and Abraham, 1979). *Telenomus* sp, *Chaetostricha* sp and *Gonatocerus* sp are some of the egg parasitoids reported (Sundararaju and Sundarababu, 2000).

Varietal tolerance

All the accessions are seen to be susceptible to the pest in varying degrees. Goa 11/6 accession exhibits certain pseudo resistance. This is by one month delay in flushing when compared to early accessions (Sundararaju, 1999). Eleven varieties were screened in Madakkathara, among these accession number 665 was the least susceptible based on shoot and panicle damage. Seedling trees are relatively less susceptible than the vegetatively propagated trees.

Seasonal pest incidence



Season-wise tea mosquito population in cashew

Based on the pest surveillance data for a period of 10 years ie, from 1988-89 to 1998-99, a graph was plotted with TMB population Vs different seasons. From the graph it is clear that the peak population build up of tea mosquito bug is in between Dec-Feb when the crop is in full bloom stage. Rapid multiplication and population build up of TMB coincides with flushing and flowering cashew which varies with the genotype of the crop and location. Hence, phenology of the crop ie, time of bud break, time of flushing under different environmental conditions is very important. Because it may some times helps to avoid the pest attack like that in case of accession Goa 11/6 which escapes the pest infestation by one month delay in flowering compared to other varieties. Here the number of bisexual and male flowers are more resulting in early fruit set and will lead to immediate thickening of the inflorescence rachii and thus avoid the attack of TMB. Pest population is totally nil during rainy season when the crop is in its dormant stage. In the field young trees are seen to be more severely attacked than the older trees. It is due to their continuous flushing nature. Seedlings are less susceptible for pest attack than the vegetatively propagated trees. This is due to the protracted flowering behaviour in vegetatively propagated trees (Beevi *et al.*, 1991). Seedling trees about 10-15 years of age is seen to be less attacked by the pest.

Inter annual variation in pest population is also noticed. For example the pest population along the west coast was very severe during 1998-99 while it was totally absent during 1997-98. Screening studies done at Madakkathara shows that accession 665 is least susceptible to the pest and the variety Dharasree is seen to be moderately resistant to the pest attack.

Influence of weather on pest incidence

The studies at Kasaragod revealed that there was no pest population from June to September when relative humidity and rainfall were quite high and period of bright sunshine was very low. The build up of pest population from a very low level (1.6%), commenced in October and reaches its peak (40.6%) in January, when the minimum temperature was very low (19.4^{0}C), minimum relative humidity was also rather low (51.6%), rain fall nil and the duration of bright sunshine was quite high (9.5 hrs.). The data also revealed that the most favourable period for rapid multiplication and pest population build up of tea mosquito was December, January and February when the mean duration of bright sunshine ranged from 9.02 to 9.8 hrs. Even though tea mosquito is known to be an insect which normally shuns bright sunlight it was observed to continue its destructive activity, hiding on the ventral surface of leaves and feeding on the mid rib, petiole, tender shoots or panicle. Moreover it is during this period that the host plant also provided an abundant supply of succulent plant parts. However, during the monsoon period, June to Sept. the relative humidity (76-82%) and rainfall (297-832 mm) were quite high, the duration of bright sunshine was too low (3.6 hrs.) and succulent plant parts were not normally available on grown up trees, and as such, the tea mosquito population was also absent (Pillai *et al.*, 1984).

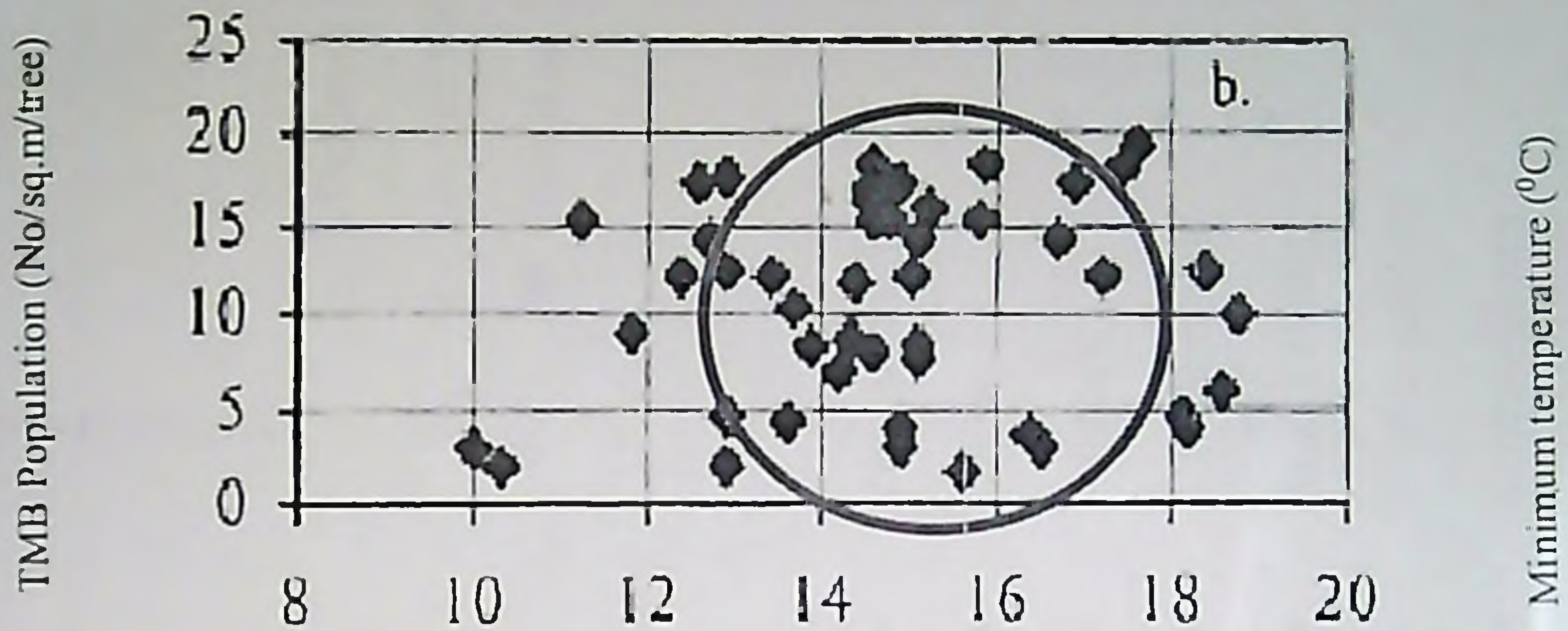
The population build up of the pest was found to be negatively correlated with the meteorological factors like minimum temperature, minimum relative humidity and rainfall and positively with sunshine. Similar was the observation made by Pilllai and Abraham, 1975 and Rai, 1981.

A comparative study at Pilicode revealed that the minimum night temperature and relative humidity in the afternoon were less during the flushing and flowering phase of cashew in 1998-99 compared to 1997-98. In a period of two and a half months, the pest might have completed three to four overlapping generations, thus increasing in number by six to seven folds causing serious damage during 1998-99 while the pest population was totally absent during 1997-98. The minimum night temperature below 20°C along with relatively dry weather in the after noon during majority of the days from December to January might have triggered pest populations during 1998-99 (Prasada Rao, 1999).

Season	Min. Temp.	Relative humidity (%)	Rainfall (mm)	Bright Sunshine hours
June-Sept	23.0	80.4	2539.9	4.4
Oct-Nov	22.4	67.6	253.6	7.2
Dec-Feb	20.3	53.3	140.9	9.4
Mar-May	24.2	64.7	498.8	8.7
Correlation Coefficient With pest population	-0.782**	-0.875**	-0.719**	0.783**

Optimum weather and pest population

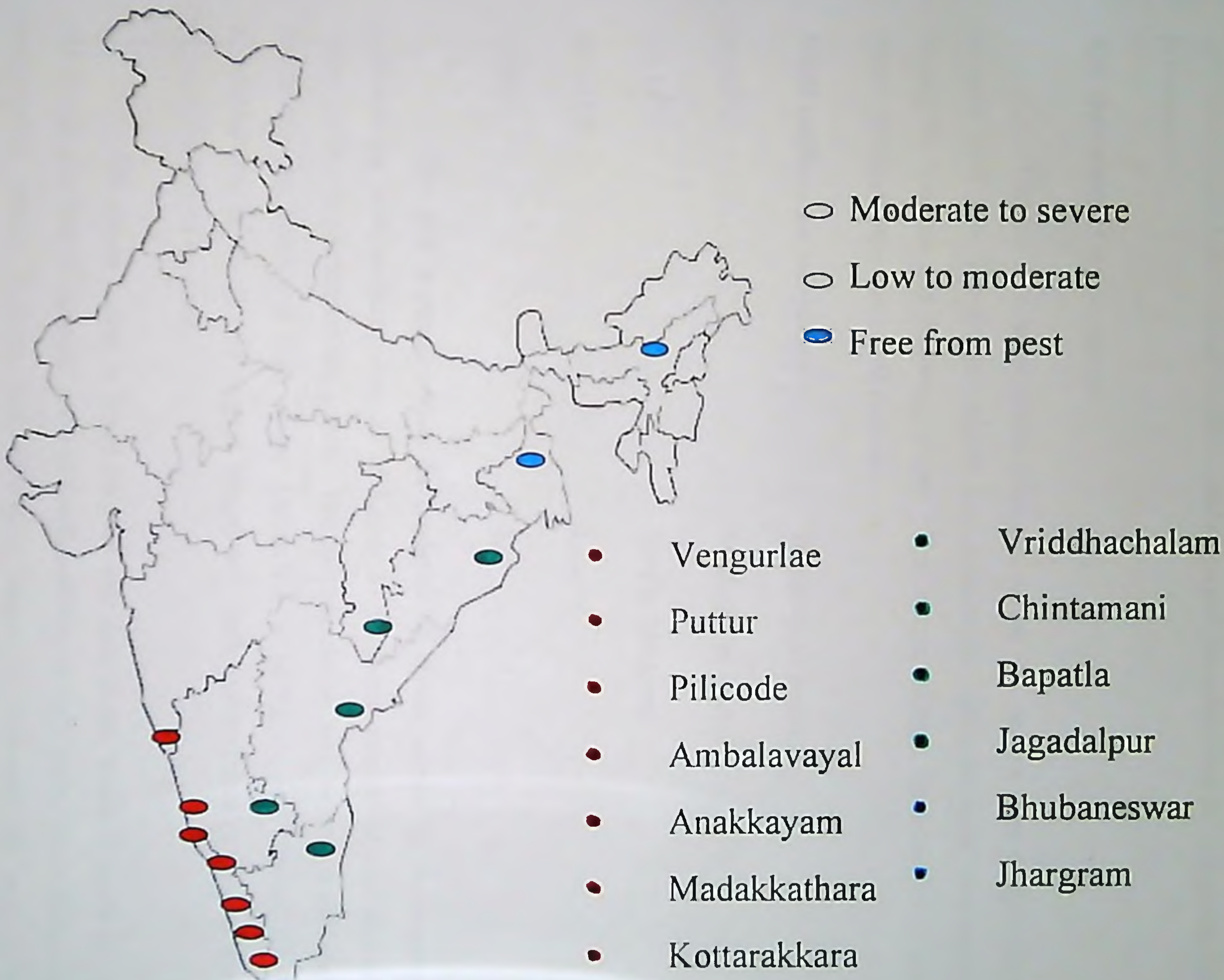
The pest population coincides with flushing and flowering phases. The time of flushing and flowering phases of cashew falls in winter (December to February) during which the night temperature plays a major role in incidence of pest population, as it is highly variable. It is also observed that minimum temperature has a significant negative relation with pest population as it is low during winter and varies between 10 and 23°C across the cashew tract of west and east coasts including "Maidan areas" of Karnataka (High ranges). The pest population was low (< 10 per sq.m.per tree) When the minimum temperature recorded below 12°C. The favourable temperature for tea mosquito incidence are 13 to 18°C.



Minimum temperature (°C) vs peak population TMB during flushing and flowering initiation (Dec-Feb) from 1988-89 to 1997-98 at ARS, Chintamani

The pest population is low under the cold weather conditions (< 12°C) during the crop season. Occasional cloudy weather may be a pre requisite and one of the important factors for pest population to build up. That is why, the pest population is high during 1990-91, 1993-94 and 1998-99 in which the bright sunshine was less due to cloudy weather. Dry weather with low relative humidity in the after noon may also favour the pest population build up as evident from path coefficient analysis. The studies at RARS, Pilicode also revealed that the minimum temperature below 20°C along with relatively dry weather in the afternoon during majority of the days from December to January might have triggered the pest population during 1998-99 across the cashew tract of Kerala (Prasada Rao, 1999). Night temperature between 13 and 18°C,cloudiness and relatively dry weather with the after noon relative humidity (40 and 60%) may be optimum for triggering pest population of tea mosquito in cashew. The cold (< 12°c of minimum temperature) hot (> 35°C of maximum temperature) and

HOT SPOT AREAS OF TEA MOSQUITO BUG



wet (continuous heavy rains) weather conditions are not conducive for triggering pest population.

Hot spot areas of tea mosquito

The hot spot areas of tea mosquito across the cashew tract of the whole country have been demarcated taking into account the optimum night temperature during the flushing and flowering stages. The criteria adopted for intensity of pest based on high temperature is as follows:-

Night surfaces air temperature	Intensity of damage
14-18°C	Moderate to Severe
12- 14°C	Low to Moderate
10 – 12°C	Low
< 10°C	Nil

The pest population may be moderate to severe across the west coast, commencing from northern districts of Kerala to Southern districts of Maharashtra where cashew is predominantly grown. The pest population may be low to moderate across the east coast and high ranges (Ambalavayal in Kerala and Chintamani in Karnataka) and relatively free in west Bengal and North eastern states as the pest may not survive in cold weather.

The above inertia is based only on two data points hence it needs to be tested and modified in endemic areas of Southern Districts of Kerala, where the night temperature during flushing and flowering stages is between 20 and 23°C (Kottarakkara and Madakkathara). Rarely the night temperature at the above places falls below 20°C during winter which coincides with flowering phase of cashew.

The potential areas of cashew production across the west coast may experience the menace of the pest. The hot spot areas of tea mosquito population need intervention through integrated pest management. However the study needs further investigations with more number of data points on pest surveillance.

Pest forecasting models

Prediction equations were derived through multiple regression analysis for each year as well as with respect to the pest population and percentage damage. The prediction equations based in the pooled data in case of pest population and percentage damage due to pest are as follows.

$Y_1 = 0.648\ x_1 + 0.002\ x_2 + 0.048x_3 - 0.073\ x_4 - 0.642\ x_5 - 0.597\ x_6 - 0.272\ x_7 + 30.473$ (R^2 - 0.53,in case of percentage damage due to pest)

$Y_2 = 1.738\ x_1 + 0.013\ x_2 + 0.138\ x_3 - 0.230\ x_4 - 1.762\ x_5 - 1.589\ x_6 - 0.791\ x_7 + 83.629$ (R^2= 0.54 in case of percentage damage due to pest)

where Y_1 = Pedicted pest population

Y_2= Percentage damage due to pest population

X_1 = Sunshine hours (hrs/day)

X_2 = Rainfall (mm)

X_3 = Morning relative humidity (%)

X_4 = Evening relative humidity (%)

X_5 = Maximum temperature (^{0}C)

X_6 = Minimum temperature (^{0}C)

X_7 = Number of rainy days

Both the regression equations explained >50% occurrence in pest population as well percent damage due to pest. However it accounted up to 90 percent in individual years. The prediction models developed by Satapathy (1993) also revealed that they explained between 80 and 90% pest population and percent damage respectively based on weather variables. The prediction equations will be useful in understanding the effects of weather elements on pest population. However the equations are not suitable for fore warming the incidence of pest.

Problems in pest forewarning

None of the institutes across the west coast is keeping data on pest surveillance which is a pre requisite to understand pest and its related environment for triggering pest population. It is evident that weather plays and important role in pest population build up. The endemic areas should be well identified with systematic collection of data on pest surveillance and weather variables. The biology of the pest under varied environments is yet to be understood under field conditions. Pest population build up coincides with reproductive phase of the crop which varies from one location to another with genotype. Information on pest environment, seasonal biology of the pest under varied environments, phenology of the crop under different field conditions etc. are required for making pest forecasting models. This should be given importance under Integrated Pest Management.

CONCLUSION

Tea mosquito bug is a menace across the cashew growing tracts of the west coast and a threat to cashew production in Kerala. Cashew production in Kerala is showing a declining trend since 1996-97 despite large scale distribution of improved planting materials and better crop management practices adopted for the last 15 years. This decline in cashew production could be reduced to a greater extent by efficient pest control measures. Cashew production in Kerala can't be sustained without pest control measures. We have seen that the population build up of tea mosquito bug is highly weather dependent. The need for pest forewarning was felt when crisis situations arises like that in 1998-99 during which there was a heavy crop loss across the west coast. None of the institute is maintaining data on pest surveillance. Unless it is taken up on priority under integrated pest management, pest warning will not be possible. If sufficient data on pest surveillance, phenology of the crop under different environmental conditions, seasonal biology of the pest under varied field conditions etc. are available, pest forecasting models can be made which helps in efficient pest forewarning. Once we can predict the pest out break in advance it will be a boon to the farming community. This helps the farmers to take necessary precautions. This approach emphasizes the need based application of pesticides and reduces environmental pollution to a greater extent.

DISCUSSION

1) Seasonal biology of the pest is important in pest fore warning how?

Peak population coincides with flushing and flowering which occurs during December-January period. Biology of the pest varies with different seasons. According to different seasons the lead time of pest population (the time period between the egg stage and the most damaging stage of the pest) varies with different seasons. Once we know the lead time of pest occurrence, prediction can be made in advance.

2) What is the relation between phenology of the crop and pest population?

Pest population build up of tea mosquito bug reaches its maximum during December-January when the trees are in full bloom stage. Time of bud break, flushing and flowering are very important in determining the pest population build up. Phenology of the crop (different life stages of the crop) varies with genotype and location. Some times the phenological characters helps the crop to escape the pest attack. Eg: in case of accession Goa 11/6, the crop escapes from pest attack by one month delay in flowering. Immediate thickening of inflorescence rachii also helps to avoid pest attack.

3) How pest forewarning is beneficial to the farmers?

Once we can predict the pest population in advance it helps the farmers to take necessary precautions. This approach finally reduces the number of sprays and leads to need based spray, which inturn strengthen the economy of the farming community as a whole. It also reduces the environmental problems to a greater extent.

4) What are the other pests of cashew?

Stem borer, leaf miner, leaf roller, leaf and blossum webber, flower thrips, apple and nut borer, hairy caterpillars and shoot tip caterpillars.

5) Why tea mosquito bug is called neem mosquito bug?

The pest was reported for the first time in tea, hence the name tea mosquito bug . Later it is seen to be causing serious damage on neem. So the pest is now known as neem mosquito bug.

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RECENT ADVANCES IN *HELICOVERPA ARMIGERA* MANAGEMENT

by

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ABSTRACT

Helicoverpa armigera is a pest of major importance in most areas where it occurs, damaging a wide variety of food, fiber, oilseeds, fodder and horticultural crops. Its biology, mobility, polyphagy, rapid and high reproductive rate and diapause make it particularly well adapted to exploit transient habitats such as man made agro ecosystem. It has been recorded feeding on 181 cultivated and uncultivated plant species belonging to 45 families (Manjunath *et.al* 1989). Its important hosts are cotton, maize, pulses, tomato, sunflower and groundnut. *H.armigera* selectively feeds on growing points and reproductive parts of their hosts, resulting in significant yield losses.

Indiscriminate use of insecticides, besides raising environmental concerns, has led to development of resistance and destruction of natural enemies. More recently, attention has been directed to bio intensive integrated pest management methods. It mainly relies on host plant resistance, biological control, cultural control and use of bio rational pesticides (Singh, 2000). The application of transgenic plants through genetic engineering is the latest concept in insect pest management (Dhaliwal and Arora, 2001).

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INTRODUCTION

Helicoverpa armigera (Hubner) is considered one of the most destructive national pests of India. It is a polyphagous pest damaging a wide variety of food, fiber, oil seeds, fodder and horticulture crops (Fitt, 1989). Among its major hosts are grain legumes, cotton, corn, sorghum, soyabean, tomato and groundnut. *H.armigera* was reported to cause $ 300 million worth of damage on legumes in India (Reed and Pawar, 1982). There are instances wherein the damage is so severe that the farmers are not even able to harvest the crops.

H. armigera has been recorded feeding on 181 cultivated and uncultivated plant species belonging to 45 families (Manjunath *et. al.*, 1989). Based on present estimates, current economic damage due to *H.armigera* largely occurs in pigeon pea (*Cajanus cajan)*, Chick pea (*Cicer artetinum*)., Tomato (*Lycopersicon esculentum),* Cotton (*Gossybium spp)*. Many other crops include sorghum, pearl millet, maize, groundnut and okra. In northern India, population of *H.armigera* is at peak on chickpea in April and on cotton Sep-Oct. The pest is reported to survive the cold winter as diapausing pupae.

H.armigera was first reported on cotton in1977 in a localized area of Punjab. In 1983 it appeared in a serious form in the northern cotton growing belt and reduced the yield by 40-50% . In south India 1987-88 in A.P& T.N and caused 66% yield loss of seed cotton.

The problem of *H.armigera* multiplied with mono cropping of cotton continuously over large areas, prolonging cotton growth beyond the regular duration, non-removal of crop residues before the next crop, the improper coverage of foliage due to the use of ineffective appliances and adoption of sub lethal dosages coupled with suspected usage of spurious insecticide aggravated the pest problems resulting in insect developing resistance to insecticides and indiscriminate use of insecticide including synthetic pyrethroids for more than 10 rounds also eliminated the natural enemies in the ecosystem.

The wide host range of *H.armigera* ensures that there is continuous availability of host plants for arvae to feed on throughout the year. In India where pigeon pea and many other host plants of *I.armigera* are grown, populations of this pests have caused severe damage to cotton leading to excessive use of insecticide. The pest has already developed resistance to *Bacillus thuringiensis*, Carbaryl, endosulfan, endrin, DDT+ Toxaphene and synthetic pyrethroids.

Use of conventional pesticides besides raising environmental concerns, has led to development of resistance and destruction of natural enemies. More recently, attention has been directed to Biointensive Integrated Pest Management. It mainly relies on host plant resistance, biological control, cultural control and use of biorational pesticides. Biorational pesticides are compounds that affect insect behaviour, growth or reproduction for suppression of insect population.

HOST PLANT AND CROP DAMAGE

For the efficient management of any pest, knowledge of its interaction with its host plants is necessary. The management of *Helicoverpa* using culturally based tactics, such as manipulation of wild hosts or cropping system, selection of resistant host plants etc. depends heavily on the basic knowledge of the relationship between the insect and its host plants.

The percentage of damage reported in various crops viz., tomato fruit borer (60-70%), chick pea pod borer (42-90%) cotton boll worm (41-56%), sorghum cob borer (40-45%), sunflower capitulum borer (29-35%), carnation bud borer (20-80%), bamboo nursery (40-45%).

The regular occurrence of *H.armigera* is limited to very few hosts, however, in certain seasons and under certain conditions, it may appear even on less preferred hosts in good numbers or in sporadic form. Its occurrence on non- preferred hosts depends on several factors such as non availability of proper physiological stage of the preferred crops, availability of succulent host, influx of migrating females or outbreak of the pest on several preferred host. On some of the generally non-preferred hosts, it gradually gets adapted and starts appearing regularly.

Among the key host plants recorded, pigeon pea was most preferred for oviposition and feeding, followed by field bean, chickpea, tomato, cotton, mungbean and sorghum. For oviposition, the host plants in the descending order of preference were pigeon pea, field bean, chickpea, tomato, cotton, mungbean and sorghum. Chilli was not a preferred host plant in free choice tests.

Besides the above mentioned host plants, *H.armigera* attacks strawberry and causes intermittent problems on pome fruits, citrus, macdamia nut, papaya, banana, etc. at flowering times and flowering ornamental plants such as gladiolus, rose, carnation, snap dragon. Serious damage by *H.armigera* in citrus orchards was observed in India in 1994-96, damaging 8.8 to 55.4 per cent fruits.

It has been reported as prominent pest in India of chicory in Himachal Pradesh; potato in Punjab and Himachal Pradesh; mustard in Satpura Plateau of Madhya Pradesh and black cumin in Himachal Pradesh (Singh *et.al.*, 2002).

Some of the weeds in appropriate succulent stage growing in and around the principal hosts of *H.armigera* support this pest, thus providing an alternative niche. In India, *H.armigera* has been recorded on about three dozen wild plants and weeds (Rao *et.al.*, 1991).

BIOECOLOGY

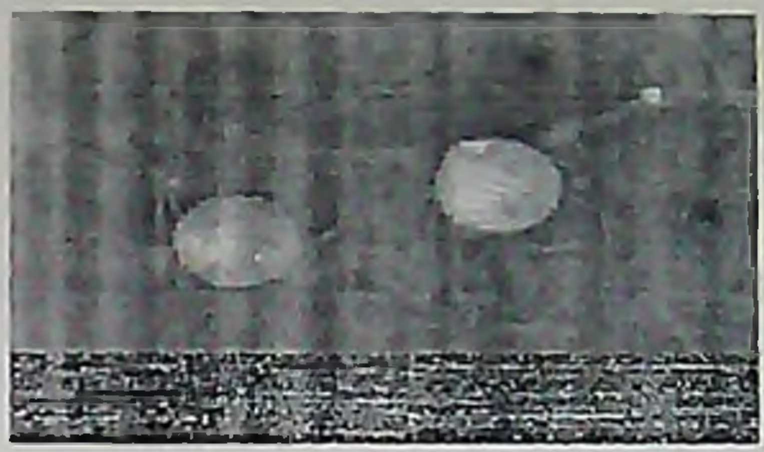
The biology of *H. armigera* on various host plants at different agro-climatic regions indicated not many differences in the developmental period, longevity etc; however, there were a lot of variations in the fecundity data. Eggs are laid singly, generally on leaves and flowers but sometimes on fruits as well. A female lays about 500 to 3000 eggs (usually more than 1000).

On hatching, the young caterpillars feed on tender foliage; advanced stage caterpillars (fourth instar onwards) attack the fruits. They bore circular holes and thrust only a part of their body inside the fruit and eat the inner contents. If the fruit is bigger in size, it is only partly damaged by the caterpillars.

Fecundity of female is high ranging between 1200 to 1600 eggs. Incubation, Caterpillar and pupal stages last for 2 to 4, 15 to 24 and 10 to 14 days respectively and the entire life cycle may be completed in 4 to 6 weeks with 5 to 8 generations in a year depending upon environmental conditions and availability of suitable hosts.

Helicoverpa armigera (Hübner, [1808])

ADULT



EGG

LARVA

PUPA

LIFE CYCLE

EGG – 2-4 DAYS
LARVA - 15-24 DAYS
PUPA - 10-14 DAYS
ADULT - 10-14 DAYS

TOTAL LIFE CYCLE - 2-4 WEEKS

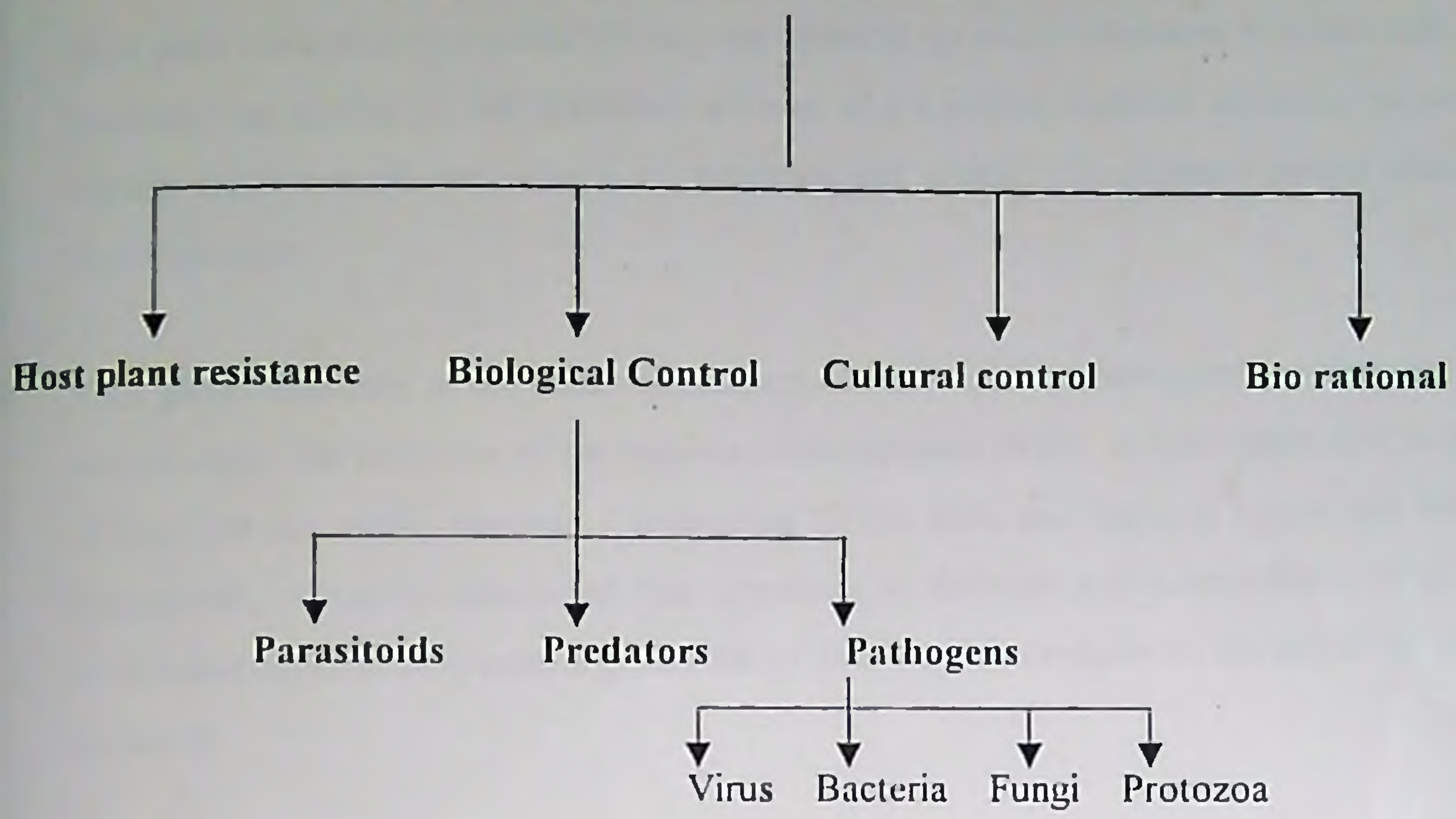
Low temperatures of 19-23^0 C and photoperiod of 11.5-12.5 h during the development of larvae are conducive for induction of diapause in the pupal stage. The diapausing pupae are located at a depth of 5-15 cm in the soil. The entry of the larvae in the soil, however, is governed by the texture of the soil and to the some extent by soil moisture. The pupae retain four pigmented larval eyespots as a characteristic of diapause. In non- diapausing pupae, such spots disappear within four days of pupation, thus serving as an excellent method of separating non-diapausing pupae from the diapausing pupae. The proportion of diapausing pupae tends to increase from zero in population near the equator to about 100% in populations from the temperate regions at the end of the favoured season.

Summer or drought responsive diapause is induced by high temperatures above 37^0 C from third instar onwards; it induced up to 94% diapause in males and 60.0% in females. In several areas of distribution of *Helicoverpa*, where the climatic conditions are optimum for development throughout the year, diapause does not occur and these pests continue to breed on their favourable hosts or weeds or some flowering plants.

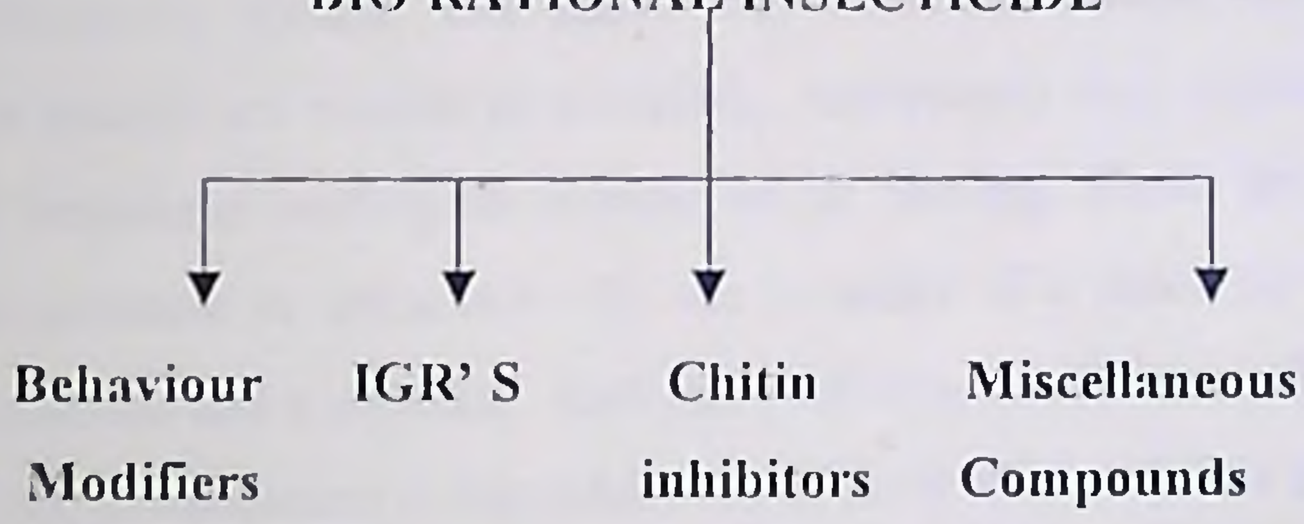
Helicoverpa are facultative migrants. There are three types of movements, i.e short range, long range and migratory, have been observed (Fitt,1989). In short range movements (100-1000m), the feeding, oviposition, mating and sheltering starts at dusk. The flight of reproductively mature moths was most frequent 1 h after sunset, the females searching for oviposition sites and nectar sources. In the second half, males are very active and undertake mate-finding flights. The majority of *H.armigera* adults dispersed below 10m height, and there was no mass ascent to higher altitudes.

Long range movements (1-10) involve less frequent responses to external stimuli and occur above the canopy at heights of up to 10 m. Adults migrate in response to poor local conditions for reproduction. Migratory movements occur above the flight boundary layer, where moths take advantages of synoptic scale wind systems at altitudes of up to 1-2 km. These flights may continue for several hours, making possible downwind displacements of hundreds of kilometers. Long flights of *H. armigera* are induced by poor larval and adult nutrition, which delay the reproductive maturity.

BIOINTENSIVE INTEGRATED PEST MANAGEMENT



BIO RATIONAL INSECTICIDE



HOST PLANT RESISTANCE

Host plant resistance represents the key for opening up major advances in insect pest management. Multilocation testing of the identified sources and breeding material needs to be strengthened to identify stable and diverse sources of resistance and identify the presence and/or emergence of new insect biotypes.

Host plant resistance is the result of interaction between two biological entities, the plant and the insects under the influence of the various environmental factor. A host plant can be resistant, more or less, but not totally immune. Considering all the flora and fauna in nature and host plant insect interactions, it can be concluded that immunity is the rule and susceptibility is an exception. In every plant species there exits a great deal of diversity with respect to the extent of damage done by an insect.

Painter grouped mechanisms of resistance into three main categories viz., non preference, antibiosis and tolerance. Antixenosis refers to the resistance mechanism employed by host plant to deter or reduce colonization by insects. The plants may deter the insects from feeding, oviposition and shelter and the insects are unable to colonises. Antixenosis may represent one or more breaks in their chain of responses leading to oviposition or feeding. These breaks may be due to (i) the absence of an arrestant or attractant (ii) the presence of a repellant (iii) an unfavorable balance between an attractant and a repellant. Antibiosis refers to the adverse effects of the host plant on the biology (survival, development or reproduction) of the insects and their progeny infesting it.

All these adverse physiological effects of permanent or temporary nature following ingestion of a plant by an insect are attributed to antibiosis. Tolerance refers to the ability of the host plant to withstand an insect population sufficient to damage severely the susceptible plants. It is generally attributed to plant vigor, regrowth of damage tissues, and resistance to lodging, ability to produce additional branches, utilization of non-vital parts by insects and compensation by growth of neighboring plants.

The *H. armigera* resistant cultivars developed for chickpea pod borer are ICC 506, ICCV 7, ICC5264, ICC6663, PDE2, ICC10667, and DULIA. ICPL 332, MA 2,PPE 45-2 are the Pigeon pea resistant cultivars. In the case of Urd, the resistant cultivars are KALAI, CO 3, CO 4, and CO 5. BT 1, T 32, T 27 are the tomato resistant cultivars. (Kalode and Sharma, 1993). Usually, a complex of allelochemicals is involved in imparting resistance to insect pests in agricultural crops. The substances like gallic, vanillic and salicylic acids, besides resorcinol, phloroglucinol and gossypol in cotton cultivars have been known to impart resistance against *H. armigera*.

CULTURAL CONTROL

Cultural practices include all the crop production and management techniques, which are utilized by the farmers to maximize their crop productivity and/or farm income. It includes decisions on crops/varieties to be grown, time and manner of planting, tillage, field and crop sanitation, application of fertilizers and irrigation, harvesting times and procedures, and even off-season operations in fallow/cropped fields.

The planting time helps to minimize pest damage by producing asynchrony between host plant and the pest or synchronizing insect pests with their natural enemies or crop production with available alternate host plants of the pest or by production followed by destruction of crop residues before the insects can enter diapause. Early sowing can be used to minimize pod borer, *H.armigera* damage to chickpea in northern India. Two peaks of *H.armigera* occur during December and March in the rabi season. During the second peak, the pest inflicts severe damage to chickpea crop. Early sown crop escapes with least damage. Late- sowing matures during late –march to aril and suffers heavy damage. November sown crop also suffers moderate damage.

Incidence of *H.armigera* on Chickpea at different dates of sowing

Sowing date	Pod damage (%)	Grain yield (kg/Ha)
October 6	1.94	2537
November 6	3.92	2373
December 6	7.70	1428
January 6	6.97	1170

Rathore and Nwanze (1993)

Inter cropping of *taramira* in *raya* reduced the incidence of mustard aphid in the latter crop due to the alleopathic influence of the former. Similarly trials conducted under the All India Coordinated Pulses Improvement project at several locations demonstrated that the sole crop of chickpea attracted more *Helicoverpa* compared to intercrops with wheat, barley, linseed, mustard and safflower (Fig.1). On the other hand, lentil and field peas as intercrops enhanced infestation in chickpea (Fig.2). Crop mixtures were more effective than row plantings (Rathore and Nwanze, 1993).

The presence of diverse vegetation within or near the field may add essential resources for predators or parasites and so enable them to find all their requirements near the pest population. Such resource includes food, cover or alternate prey. Conversely, weeds may also adversely affect the orientation of predators and parasites to their prey. The weeds may even directly contribute to pest multiplication by providing preferred surface for oviposition. *Vicia sativa* is a common weed associated with chickpea in northern India. Removal of the weed at a time when maximum eggs are laid substantially reduces the incidence of pod borer, *H.armigera.*

Effect of weed removal on the incidence of *Helicoverpa armigera* (Hubner) in chickpea

Year	Pod damage (%)		Reduction in infestation (%)
	Control	Weed removed	
1984-85	64.70	16.00	75.20
1985-86	33.40	21.10	36.90
1986-87	50.40	31.90	36.80

Trap cropping of marigold after every 8 rows of tomato attracts most of ovipositing moths to the former crop. The use of conventional insecticides on the trap crop reduces their attractiveness to the pest. Therefore, the pest on the trap crop has to be removed mechanically. The residual pest population on both the crop is controlled by sprays of *H.armigera* nuclear polyhedrosis virus (HaNPV) at 500 larval equivalents (LE).

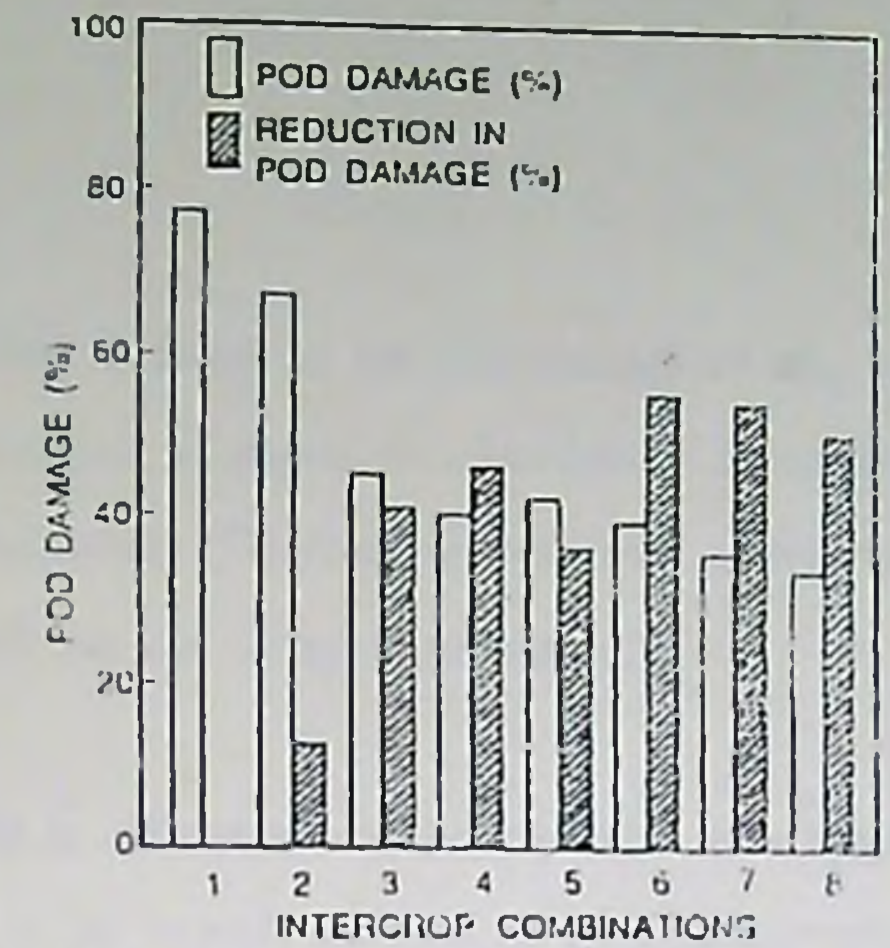


Fig. 1 Incidence of *Helicoverpa armigera* (Hubner) in chickpea sown alone and as inter/mixed crop. 1 = Chickpea (sole) ; 2 = Chickpea (sole-paired planting) ; 3 = Chickpea + Wheat (2 : 1) ; 4 = Chickpea + Barley (2 : 1) ; 5 = Chickpea + *Raya* (4 : 1) ; 6 = Chickpea + Wheat (mixture) ; 7 = Chickpea + Barley (mixture); 8 = Chickpea + *Raya* (mixture) (After Rathore and Nwanze, 1993). Incidence on chickpea (sole) is higher as compared to intercropping with wheat, barley and *raya*.

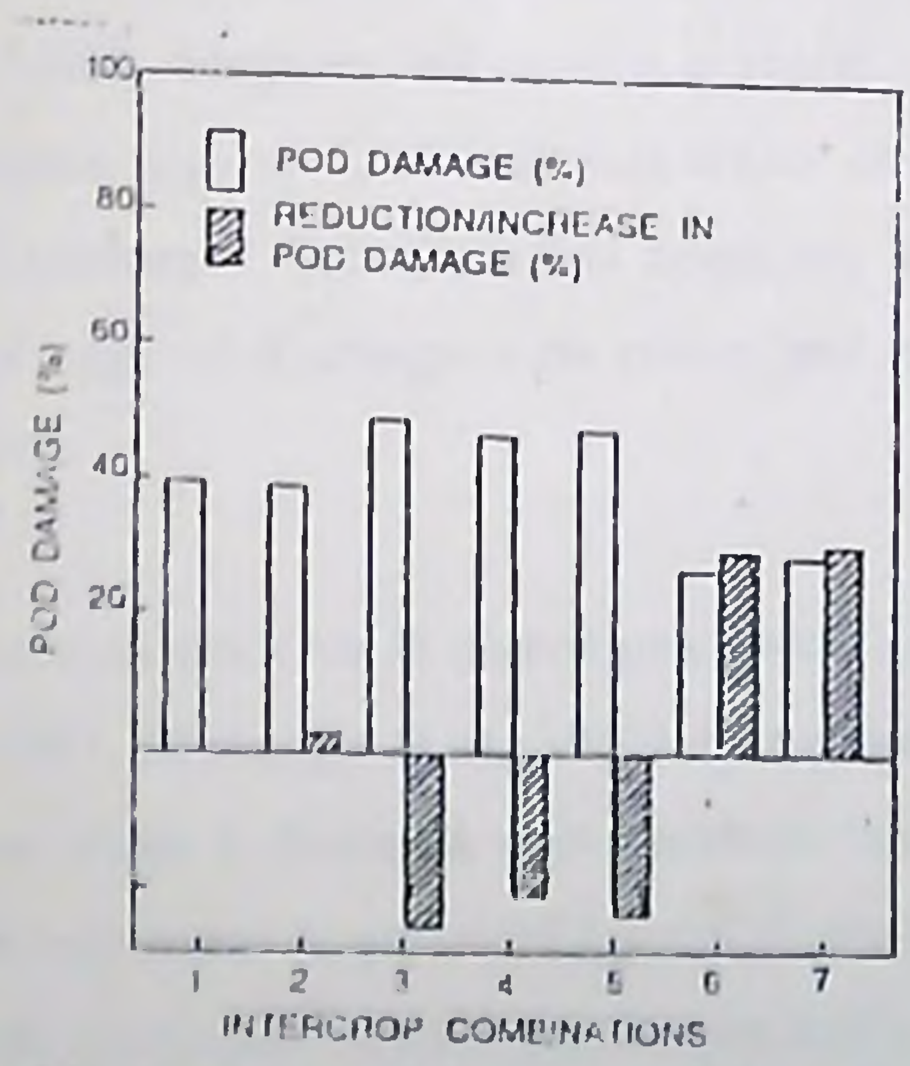


Fig. 2 Influence of intercropping on pod damage due to *Helicoverpa armigera* (Hubner). 1 = Chickea (sole) ; 2 = Chickpea (sole paired) ; 3 = Chickpea + Field pea (2 : 3) ; 4 = Chickpea + Field pea (2 : 2) ; 5 = Chickpea + Lentil (2 : 2) ; 6 = Chickpea + Bakla (2 : 2) ; 7 = Chickpea + Linseed (2 : 2) (After Rathore and Nwanze, 1993). Intercropping with field pea (3, 4) and lentil (5) reduced incidence of *H. armigera*.

Maize and sorghum on the borders of pigeon pea acts as bird perches that eat pod borer larvae and also harbors natural enemies of pigeon pea pests.

BIOLOGICAL CONTROL

Parasitoids

Out of the 77 parasites recorded in India so far (Manjunath *et al.*, 1989), fifty belong to parasitic *Hymenoptera, Banchopsis ruficornis, Campoletis chlorideae, Enicospilus* sp., *Eriborus* sp., *Bracon brevicornis, Trichogramma chilonis, Trichogrammatoidea armigera, Palexoristalaxa, Carcelia illota and Goniophthalmus halli* are the common parasitoids.

Parasitism by trichogrammatids is influenced by host plants. The eggs of *H.armigera* on marigold, tuberose and corn were found to be heavily parasitised by *Trichogramma Chilonis* (up to 85%). On tomato and niger, 80% and 20% parasitism by *Trichogramma* was found. The eggs from tomato Lucerne and potato, particularly potato, were parasitised heavily by *T. chilonis* (up to 84.2-98.2%) (Yadav and Patel, 1981).

In Sorghum, cotton, cowpea, mungbean, groundnut, corn, sorghum and pearl millet, up to 85% parasitism has been recorded. Sorghum and cowpea attracted maximum egg parasitism. In cotton, parasitisation of *H.armigera* eggs by *T. chilonis* was lowest on *Gossypium raimondii* (25.3%), and highest (57.30%) on *G.harknessii*. Parasitism was negatively associated with trichome density. In Karnataka, parasitism of eggs of *H.armigera* on cotton and intercrop okra (trap crop for *Earias* spp.) was 51.16 and 4.84%

Due to the acidic exudates on chickpea & pigeon pea plants, parasitism was either nil or extremely low (Yadav and Patel, 1981; Bhatnagar *et al.*, 1982). In Andhra Pradesh, *Trichogramma* was more abundant on pigeon pea when it flowered after sorghum than when it flowered with sorghum. Hence the failure of *Trichogramma* to parasitise eggs of *H.armigera* on the pigeon pea was not due to the failure of the adult parasitoids to enter pigeon pea fields, but due to some other unexplained mechanism. Studies conducted in India have revealed that *T. chilonis* can parasitise up to 34% of *H.armigera* eggs if they are laid on the vegetative stages of pigeon pea crops.

Only 0.1% of eggs of *H.armigera* on pigeon pea were parasitised by *Trichogramma* (Bhatnagar *et. al.*,1982). However, 7.8%-69.2% parasitism was observed on short duration pigeon pea intercropped with sorghum. Parasitism was high and concentrated on the first flush of pigeon pea flowers and declined on the second flush of flowers.

Among the different larval parasitoids on *H.armigera, C.chlorideae* and *C.illota* were most active on seasonal crops and also during most seasons. Manjunath et *al.*, 1989 have provided information on the influence of seasons and host plants on parasitism of *H.armigera* by various species. Hymenopterans were predominant on sorghum and chickpea and dipterans on pigeon pea.

Parasitism by dipterans on sorghum, pigeon pea and chickpea was up to 2.1%, 22% and 3% respectively and by hymenopterans on sorghum, pigeon pea and chickpea was 4.9%, 5% and 17.2% respectively (Bhatnagar *et. al.*, 1982). Recent search for parasitoids carried out in nine Indian states in sprayed cotton fields resulted in the record of egg parasitoid *T. chilonis* from four states and five places. Early larval parasitoids *E. argenteopilosus, C.chlorideae* and *Apanteles* sp. were recorded from three states with maximum numbers from Akola, Maharastra.

Campoletis chlorideae is most common and widely distributed from Himalaya to southern India. It parasitises its host on cotton, chickpea, sunflower and many other crops, parasitism ranging from 35 to as high as 83%. In Andhra Pradesh, Karnataka and Maharastra *C.chlorideae* is the most effective parasitoid of *H.armigera.* It is widespread and found at all times of the year on a wide variety of hosts. Parasitism is generally highest in September and lowest in May. At one site during 1977-83, average parasitism by *C.chlorideae* was 44.2,33.1, 32.6,7.1 and 4.2%, respectively, on sorghum, chickpea, pearl millet, groundnut and pigeon pea. Ten species of hyper parasitoids were recorded from cocoons of *C.chlorideae* with hyper parasitism being about 40% in cereals and 10% in legumes (Pawar *et al.*, 1989).

PREDATORS

Predators such as *Delta, Orius, Chrysoperla, Cheilomenes, Rhynocoris, Geocoris, Nabis,* carabids, ants, mantids, spiders, and birds feed on *H.armigera* eggs, larvae, prepupae and pupae. The role of predators in regulating *Helicoverpa/Heliothis* populations has not been quantified. But in India, the wasps *Delta spp.* and *Chrysoperla* spp. have been observed to be important predators of *H.armigera*. Predation of *H.armigera* eggs by second instar larvae of *C.carnea* was highest on *Gossipium arboreum*, MCU 9 and *G. harknessii*, each predator consumed 18.33, 18.00 and 17.33 eggs/day, respectively.

Ghode have recorded that in chickpea field in Orissa during the 3rd week of January 1987, due to the presence of birds, the population of *H.armigera* was reduced from 5-10 larvae/plant in mid January to a trace by the end of the month. In pigeon pea ecosystem, black drongo, house sparrow, green bee-eater and blue jay accounted for 60.3, 19.8, 12.9 and 6.8 percent avian population, respectively. The predatory birds cleared 56% larvae from inanimate perches and 26% from animate perches. Late instar larvae were picked in large proportion (80%) by black drongo within a radius of eight meters from inanimate perch and two meters from animate perch.

Orius is considered to be one of the most important natural enemies in a number of ecological zones. The house sparrow (*Passer domesticus*) was the most important bird predators of larvae and adults of *H.armigera* on Egyptian clover in Punjab and on chickpea, *P.domesticus* and black myna (*Acridotheres ginginianus*) were the important predators. Ants have also been found to be potential predators in sunflower and pigeon pea ecosystems in Karnataka (Singh *et.al.*, 2002)

PATHOGENS

Natural epizootics of nuclear polyhedrosis virus (*Ha*NPV) in larvae on corn and cotton have been recorded in Uganda. A virulent strain was identified in Ivory Coast. Higher incidence on corn than cotton was recorded. *Ha*NPV was most frequently on *H.armigera* larvae collected from cleome and corn in Western Tanzania. In India the populations of 3rd to 6th instar larvae of *H.armigera* declined by 22.75 to 23.08 owing to infection by *Ha*NPV. In China, the natural infection rates of *H.armigera* moths collected in light traps reached 84.9%.

A granulosis virus and a cytoplasmic polyhedrosis virus have been recorded from *H.armigera* from India. A gonad specific virus from *H.armigera* has been suspected based on the external manifestation of waxy-plug symptom of the genital system.

Among the bacterial Pathogens, the records include *Bacillus thuringiensis* Berliner from India, *B. thuringiensis* subsp. *kurstaki*, *B. thuringiensis* var. *galleriae* Krieg and *Serratia marcescens* Bizio from Russia , B.*cereus* and *B. thuringiensis* from China and Egypt. Several subspecies of *B. thuringiensis*, namely *aizawai, dendrolimus, entomocidus, galleriae, kurstaki, shandonggensis*, and *thuringiensis*, have been recorded on *H.armigera* in different areas of its distribution.

The important fungal diseases isolated from *H.armigera* include *Beauveria bassiana* Vuillemin and *B.brongniartii*, *Metarhizium anisopliae* Sorokin, *Nomuraea rileyi* Samson from India, *Fusarium oxysporium* var *orthoceras, Aspergillus parasiticus* Sphere and *Scopulariopsis brevicaulis* Bianer from Russia, *M. anisopliae, B. bassiana* and *N. rileyi* from Indonesia, and *B. bassiana* from Charles and *Entomophthora* sp. have also been recorded.

Beauveria sp. was found infecting *H.armigera* on cotton, pigeon pea and other legumes in Andhra Pradesh in October-December, 1987. Naturally occurring epizootics by *N. rileyi* have been noted in cotton, corn, sorghum, soybean and other crops.

Epizootology studies of *N. rileyi* in field populations of *H.armigera* in India have revealed higher rates of fungal infection in *H.armigera* on *Cajanus cajan* (37%) compared to *Phaseolus vulgaris* (28.2%) and tomato (20.5%). In the field, 5-8 day old larvae attacked by *N. rileyi* give rise to adults and only 5% did so when they were first attacked when 11 days old (Gopalakrishnan and Narayanan, 1989).

In India, in 1998 when outbreak of *H.armigera* occurred on cotton and chickpea, naturally occurring epizootics by *N. rileyi* was recorded. More than 35% of the larvae infected yielded mycelium and spores of *N. rileyi* and only about 6% yield white muscardine fungus, *Beauveria bassiana*. The availability of *H.armigera* and *Spodoptera litura* in exceptionally large numbers and favourable climatic conditions appeared to have triggered the infection.

Larval diseases of *H.armigera* such as nuclear polyhedrosis, *N. rileyi* and *B. bassiana* were recorded from all the nine Indian states recently surveyed.

In general, entomopathogenic fungi have their greatest potential as microbial agents in situations where the microclimate in the area of the host provides high humidity with temperatures between 20 and 30^0 C and where moderate feeding by pest larvae prior to death can be tolerated. Epizootics typically occur when host densities are high late in the season, after the need for plant protection has passed, but these could be usefully employed area wide to reduce the overall pest population.

Among the protozoans, *Nosema* sp, *Vairimorpha* sp. and *Mattesia* sp have been recorded from India and *N. medinalis, V. necatrix* Kramer from China. Additionally, *N.heliothidis Plistophora* sp and *Thelohania* sp have also been reported from *H.armigera* from time to time.

Ovomermis albicans, Hexamermis spp. and *Pentatomimermis* sp from *H.armigera, H.assulta* and *H.peltigera, Heterorhabditis bacteriophora* Poinar, *Steinernema* spp and *Steinernema feltiae* have been recorded from India. Entomopathogenic nematodes have been recorded from many distribution zones of *H.armigera* nematodes are generally more active on altisols than vertisols.

In India, Bangalore, during the rainy season. The nematode *Hexamermis* sp was very active on tomato crop and *O.albicans* was recovered from *H.armigera, H.peltigera* and *H.assulta* infesting various crops and weeds in Andhra Pradesh. Parasitism by *O.albicious* was found to be higher on groundnut, tomato and weeds.

A number of trials have been conducted in India (Tamil Nadu), to check the efficacy of *H.armigera* nuclear polyhedrosis virus(*Ha*NPV) for the management of the pest on different crop plants

Jayaraj(1990) observed that application of *Ha*NPV with 20% starch or 1% sugar provided good UV protection and that *Ha*NPV was more effective on chickpea than on lablab. Adjuvants and UV protections like Ranipal(0.5%), robin blue(0.5%), Tinopal(1%), crude sugar, egg homogenate, cottonseed oil, etc with *Ha*NPV when sprayed on chickpea could reduce pod damage and increase yield. The persistence of *Ha*NPV could also be increased. *Ha*NPV was more effective on highly susceptible and moderately susceptible chickpea varieties (Shoba and Annigeri) in comparison to tolerant varieties (ICC 506 and ICC 10817) (Rabindra *et al.*, 1992).

For the management of *H.armigera* on pigeon pea, ultra low volume and wettable powder formulations of *Ha*NPV were found to be as effective as endosulfan. *Ha*NPV could cause 40-77.9% mortality of *H.armigera* on sunflower. On groundnut, a single application of NPVs of *H.armigera* at 250 LE (1.5 X 10^{12} POB), with crude sugar was as effective as two applications of Chlorpyriphos (Dhandapani *et. al* .,1993).

Two applications of *Ha*NPV at 250 LE /ha at 7 and 10 days intervals were very effective in reducing the larval population and increasing grain yield in 5 sorghum varieties in Tamil Nadu (Dhandapani *et.al*.,1993). In Gujarat, five sprays of *Ha*NPV at 250 LE/ha per week gave satisfactory control of *H.armigera* and resulted in a grain yield increase of 28% - 47% in chickpea.

In Maharastra, two applications of *Ha*NPV @ 450 LE/ha at 10 day interval were most effective (Dhamdhere and Khaire, 1986). However, in further studies, two sparys of *Ha*NPV @ 500 LE/ha were as effective as two sprays of endosulfan and gave higher yields(Pawar *et al*., 1989). When Sorghum was treated with *Ha*NPV, the lowest cost benefit ratio was obtained. In Karnataka, *Ha*NPV @ 500 LE/ha together with either 0.1% boric acid or 0.5 % jaggery was effective in managing *H.armigera* on sunflower and red gram (Bijjur *et. al*., 1991).

In on farm field experiments at 5 sites in Karnataka on tomatoes, spraying with a formulation of *Ha*NPV against *H.armigera* significantly decreased larval count and increased fruit yield (Gopalakrishnan and Asokan, 1998). *Ha*NPV @ 250 LE/ha and a combination of *Bacillus thuringiensis var.kurstaki* 1 kg + *Ha*NPV 125 LE/ha were equally effective in chickpea ecosystem. *H. armigera* on tomatoes was controlled by Dipel at 0.5 kg/ha sprayed every 10 days, which had the same effect as fortnightly applications of monocrotophos 0.05 kg/ha (Krishnaiah *et al* ., 1981).

When *Beauveria bassiana* @ 2.68 x 16^{7} spores/ml was applied, the average chickpea pod damage was 6% and the yield was 2377 kg/ha. The untreated control recorded 16.3% pod damage with a yield of 1844 kg/ha. In preliminary experiments conducted in India (Bangalore), *Steinernema bicornutum* proved effective against *H.armigera*. It was found to be compatible with nuclear polyhedrosis virus of *H.armigera* and pesticides such as endosulfan, some synthetic pyrethroids and botanicals. It was also found to be safe to predatory birds like black drongo and target specific

BIORATIONAL INSECTICIDES

Behaviour modifying chemicals

Insect behaviour is elicited in response to visual, auditory, tactile, olfactory and gustatory- sensory information from the surrounding world. However chemical communication appears to be the primary mode of information transfer in most groups of insects. Chemicals involved in communications are termed as Semio chemicals and divided into interspecific and intraspecific communication chemical. Pheromone is defined as a chemical or a mixture of chemical that is released to the exterior by an organism and causes one or more specific reactions in a receiving organism of the same species. Sex Pheromone is a substance generally produced by the female to attract males for the purpose of mating. The sex pheromone of *H.armigera* contain Z11-16: Ald, Z9-16 Ald 16 and 16: OH. They also reported that Z11-16: Ald in combination with 1-10 percent Z9-16 Ald proved to be an effective lure for *H.armigera* in Israel (Dunkelblum *et al.*, 1980).

Sex Pheromones of Cotton bollworms have been extremely useful for monitoring, survey and surveillance and assessing damage in cotton in India. In Punjab, Pheromone traps are used for monitoring for the activity of *H.armigera*. Monitoring the population of Boll worms can help in their early detection and build up and a need based control measure can be adopted depending upon the severity of a particular species of Boll worms.

Chitin synthesis inhibitors

Chitin synthesis inhibitors are the compounds, which inhibits the synthesis of chitin. Foliar applications of diflubenzuron have shown excellent residual activity against eggs of *H.armigera*. In addition to its well-known stomach toxicity, it was found to have significant contact activity against *H.armigera*.

Spinosyns

Spinosyns are perhaps the newest class of insecticides, represented by spinosad (Success®, Tracer Naturalyte®). Spinosad is a fermentation metabolite of the actinomycete *Saccharopolyspora spinosa*, a soil-inhabiting microorganism. It has a novel molecular structure and mode of action that provide excellent crop protection typically associated with synthetic insecticides, first registered for use on cotton in 1997. Spinosad is a mixture of spinosyns A and D (thus its name, spinosad). It is particularly effective as a broad-spectrum material for most caterpillar pests at the astonishing rates of 0.04 to 0.09 pound of active ingredient (18 to 40 grams) per acre. It has low mammalian toxicity and selectively active on Lepidopteran pests. It has also been reported to be highly effective against *H. armigera* on chick pea and on cotton.

BIOTECHNOLOGICAL APPROACHES

Existence of complex and dynamic interactions between populations of pests and entomophagous influenced by physical and biotic factors makes it difficult to predict an entomophagous arthropods ability to reduce pest levels. However, there are various criteria for choosing a potential biotic agent, like adaptation to the abiotic environment, appropriate host range, good searching ability, synchrony with host life cycle, pesticide tolerance, ease of rearing, etc. Different strains of biotic agents can be collected from different agro-climatic regions or from different situations. The desirable and undesirable characteristics of these strains are determined to select the potentially good candidates for augmentation.

In China, two different strains of *T.chilonis* were collected from eggs of the noctuid *Anomis flava* (Fabricius) on cotton and the tortricid *Acleris(=Peronea) crocopepla*(Meyrick) on apple trees and were released on cotton, rice and pepper for the control of *H.armigera* and *H.assulta*. The fruit strain appeared to be more efficient in controlling the pest (Huo *et. al.*, 1988)

Different species and strains of *Trichogramma* were tested against *H.armigera* (Hassan, 1988). It was found that *T.dendrolimi, T.evanescens and T.chilonis* were the most effective.

Genetic variability in host infestation efficacy was studied in old and new strains of *Trichogramma bourarchae* Pintureau and Babault in France. The old strain consisted of one line collected in 1988 from *Cynthia cardui* on mallow. The recently captured line had a higher parasitization capacity. Artificial host eggs have been manufactured for rearing *Trichogramma* in China. *T.dendrolin* reared on artifical eggs was released in cotton field @ 1,50,000 parasitoids for the control of *H. armigera*. The searching and parasitising ability of these parasitoids were as good as those obtained from normal eggs (Dai *et al.*, 1988).

T. chilonis from cotton ecosystem of six different agro-climatic zones of India were compared for 40 generations. The Gujarat and Punjab strains named as BioC1 and BioC2 respectively, were superior to other ecotypes(Jalali and Singh, 1993).

For geographical strains of *C.chlorideae* were obtained from four different states in India and their performance compared (Ballal and Ramani,1994). Sehore (Madyapradesh) and Rahuri (Maharastra) strains were found to be superior with the former proving to be the most fecund with better searching ability.

Five entomogeneous fungi were bioassayed for their infectivity to second instar larvae of *H.armigera* in India. *B.bassiana* (Bapatla isolate) was found to be the most virulent (Prasad *et al.*, 1990).

Seventeen strains of *Bacillus thuringiensis* were assayed against larvae of *H.armigera*. Strains producing CryIA (c) were more toxic than CryIA (b), CryIB, CryIC and CryIC and CryID. *B.thuringiensis* subsp. *kurstaki* HD-73 was the most toxic.

Comparative studies of replication and infectivity of *Ha*NPV conducted in China showed that the virus replicated much faster in heterologous cells than in the homologous cells. In Turkmenistan, Agalykov *et. al.*, (1990) have developed a method for obtaining highly pure virulent polyhedra from infected larvae of *H.armigera*. Hanzlik *et. al.* (1993) isolated a small RNA virus from *H.armigera*, which was called the *H.armigera* stunt virus. It caused severe retardation of the development of *H.armigera* and led to subsequent death.

In a green house trial in Belgium, significant control of *H.armigera* was observed on transgenic tomatoes expressing *Bacillus thuringiensis* toxin. Percentage fruit injury and number and size of larvae were reduced in transgenic plants (Jansens *et. al.*, 1992).

A virulent gene product, known as viral enhancing factor, has been identified by Granados and Corsaro (1990) that could enhance infection 25-fold. This factor was identified in *H.armigera* granulosis virus and it is considered to have the potential for genetic improvement of viruses.

The invasion rate was determined for 2 strains of *Steinernema carpocapsae (Neoaplectana carpocapsae)* and one strain of *S.glaseri (N.glaseri), Heterorhabditis bacteriophora and Heterorhabditis* sp. On 4th instar larvae of *H.armigera*. The LT $_{50}$ for *H.bacteriophora* were 6-30 times as high as those of the other nematodes. Insect susceptibility and nematode infectivity as measured by the invasion rate assay were compatible with the results from the dose response assay (Glazer, 1992).

TRANSGENIC CROPS

Transgenic crops expressing insecticidal proteins have considerable promise for use in IPM programmes against *H.armigera* and other lepidopterous pests. Genes expressing Bt toxins have been successfully introduced in crops like cotton, corn, pigeonpea, chickpea, sorghum, groundnut, tomato, etc. Significant variation in insecticidal activity has been observed during different growth stages and different plant parts of these crops.

Selvapandian *et.al.*, (1998) have succesfuuly transformed tobacco and chickpea cultivars, respectively using Bt toxin gene in India that provided complete protection against *H.armigera*. Efforts are also on to develop transgenics in sorghum, pigeon pea, and chickpea to control *H.armigera* at ICRISAT. Such transgenic crops are extensively being cultivated in the USA, Australia, and China etc. The Government of India too has approved the use of transgenic cotton (Bollgard®) from 2002 with certain conditions like provision of refugia. Varieties released for cultivation in India are MECH – 12 B.t, MECH – 162 B.t and MECH – 184 B.t.

Recent studies on the delta-endotoxin structure show that it has three domains.

Domain I is a bundle of 7 alpha helices, some or all of which can insert into the gut cell membrane, creating a pore through which ions can pass freely.

Domain II consists of three antiparallel beta-sheets, similar to the antigen-binding regions of immunoglobulins, suggesting that this domain binds to receptors in the gut.

Domain III is a tightly packed beta-sandwich, which is thought to protect the exposed end (C-terminus) of the active toxin, preventing further cleavage by gut proteases. Interestingly, the diphtheria toxin (of another bacterium) has an essentially similar structure to the Bt toxin.

BIOINTENSIVE INTEGRATED PEST MANAGEMENT FOR DIFFERENT CROPS

BIPM for *H.armigera* in Red gram

a) Spray NSKE 5% + 1% soap solution or neem oil/kernel based formulation as Oviposition deterrent.

b) Grow tall sorghum as companion crop to serve as biological bird perches.

c) NPV 1.5 X 10 ^{12}/ha three times starting from flowering stage at 15 days interval.

d) Apply B.t.k. @ 600 unit in place of NPV if it is most effective.

e) Use spray adjuvant: Teepol 0.1% or jaggery 0.5%

f) Set up pheromone trap, ETL 5-6 moths/trap.

BIPM for *H.armigera* in Sunflower

a) Release *Chrysoperla* @ 1 per bud

b) Apply NPV at 1.5 x 10 ^{12} POB/ha coinciding with anthesis

c) Apply endosulfan 0.07% if other pets are noticed or for H.armigera after seed settling.

BIPM for *H.armigera* in Tobacco

a) Grow *Nicotiana rustica* as ovipositional traps in two rows around the main tobacco

b) Apply Tobacco decoction as an ovicide on trap crop during peak oviposition

c) Apply NPV 6.0 X 10 ^{12} POB/ha 70.25% boric/tannic acid three times at weekly intervals, first commencing from flowering stage.

BIPM for *H.armigera* in Tomato

a) Six releases of *Trichogramma chilonis* @ 50,000/ha/week, first release coinciding with flowering time and based on ETL of 4-6 moths/six pheromone traps.

b) Apply NPV @ 1.5 X 10 ^{12} POB + Cotton seed kernel 300 g/ha thrice. Each application should be followed by Trichogramma release.

c) Grow trap crop: Plant simultaneously 40 days old African tall marigold and 25 days old tomato seedling (1:16 rows)

CONCLUSION

There is an ocean of information available on various aspects of *H. armigera*, yet there is need to conduct exhaustive studies on the following lines:

1. Determine parasitoids /host plant relationship in all the distribution zones of *Helicoverpa*
2. Develop cost- effective mass production system.
3. Identify constrains in utilizing bio control agents.
4. Evaluate the economics of augmentation in most promising situation.
5. Develop novel methods of transfer of viable technologies, invclving farmers.
6. Study the feasibility of use of microbials to replace some of the insecticides.
7. Breed predator/ parasitoids friendly varieties.
8. Use transgenic crops expressing insecticidal protein.

For success of any biological control programme, national and international cooperation is essential. A network on BIPM of *Helicoverpa* has to be developed for all host plant.

DISCUSSION

a. What is the difference between LE and POB?

One Larval Equivalent is $6x10^9$ POB/ml. Previously the field dose is recommended in Larval Equivalent now it is changed to number of Poly Occlusion Bodies per mill liter.

b. What are all the commercial formulations on B.t and NPV available in India?

In the case of virus the commercial formulations are Helicide and Elcar. Thuricide, Biolep, Bioassp,Delfin,Dipel are the *Bacillus thuringiensis* formulations available in India.

c. Who has given approval for the release of B.t cotton?

The approval for the commercial release of Mahyco's B.t – cotton was given by the Genetic Engineering Approval Committee (GEAC) of the Ministry of Environment and forests, the statutory body set up for approving large-scale use of genetically engineered organisms.

d. Is there any report on transgenic B.t cotton developed resistance?

No, there is no possibility of developing resistance but the gene may get silenced.

e. What are all the traditional practices available for the management of *H. armigera*?

Trap cropping of marigold after every 8 rows of tomato attracts most of ovipositing moths to the former crop. Maize and sorghum on the borders of pigeon pea acts as bird perches that eat pod borer larvae and also harbors natural enemies of pigeon pea pests.

f. Suggest some resistant management practices for *H.armigera* ?

Set up pheromone traps to monitor the level of infestation. If it exceeds ETL go for insecticide application. Avoid the use of resurgence causing chemicals. Alternate the chemical insecticide application with bio control agents.

g. What are all the chemical the pest has developed resistance ?

The pest has developed resistance to Carbaryl, endrin, DDT+ Toxaphene and synthetic pyrethroids.

h. What about Diapause in *H.armigera*?

H.armigera undergoes diapause in pupal stage. Low temperatures of 19-23^0 C and photoperiod of 11.5-12.5 h during the development of larvae are conducive for induction of diapause in the pupal stage

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USE OF DIFFERENT TRAPS AND LURE MATERIALS FOR THE FRUIT FLY MANAGEMENT

SEMINAR REPORT

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ABSTRACT

Fruit flies play havoc with horticultural crops. They cause tremendous economic loss and great problem to fruit and vegetable growers all over the world. The estimated annual loss due to fruit fly attack is around Rs. 2,600 crores in India (Stonehouse, 2001). In the management of fruit flies, behaviour-manipulating strategies like use of traps and attractant materials are gaining importance as an alternate to chemical control measures.

Common types of traps used in the capture of fruit flies are Steiner trap, McPhail trap, Jackson trap, Bucket trap, GK trap etc. (Cornelius *et al.,* 1999). Lure materials used for fruit fly attraction can be categorised into parapheromone lures, food lures and visual cues. Vargas *et al.* (2000) reported that bucket traps baited with the parapheromone, cue-lure was very effective in capturing the males of melon fly, *Bactrocera cucurbitae* (Coq.) and the weekly catches varied from 543.3 to 803.8 per trap. McPhail traps baited with the food lure, protein hydrolysate was found to be more attractive to the Oriental fruit fly *B. dorsalis* (Hendel) when compared to the other baits like fermented palm juice, sugar and ripe mango pulp. (Agarwal and Kumar, 1999)

USE OF DIFFERENT TRAPS AND LURE MATERIALS FOR THE FRUIT FLY MANAGEMENT

Introduction

Fruit flies play havoc with horticultural crops. They cause tremendous economic losses and great problems to fruit and vegetable growers all over the world. They attack a wide variety of vegetables like cucurbitaceous crops, tomato, brinjal and fruit crops like mango, guava, sapota, papaya etc. Fruit flies attack the crop in its most vulnerable stage and typically cause yield losses to the tune of 10 to 40 per cent in India and the estimated annual losses in around Rs.2,600 crores (Stonehouse, 2001).

The strategy for the control of insect pests in fruit and vegetable crops in necessarily to be different from others, because of the nature of utilisation. This calls for extra attention, especially in case of chemical insecticides, because of hazardous residues of insecticides. The other problems like insecticide resistance, resurgence, and adverse effect on natural enemies also warrant the use of insecticides. Hence, an eco-friendly approach is necessary for the management of pests of fruits and vegetables. Behaviour manipulating strategies like use of insect traps and attractant material are now days widely used for the management of fruit flies.

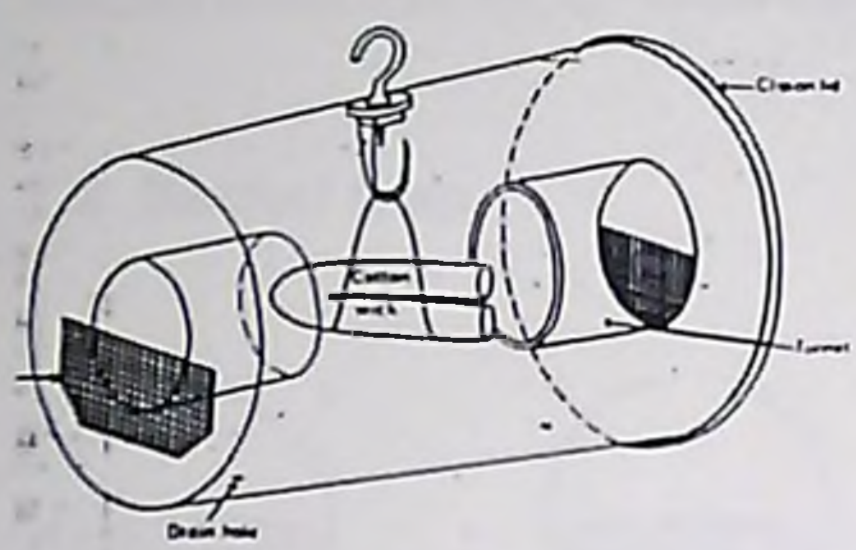
The unique behavioural adaptation of the fruit flies also renders the pest management program very difficult. The female fruit fly using long ovipositor by the eggs beneath the fruit skin about 3-5 mm deep inside. Developing larvae are also seen inside the fruit and are hence inaccessible. The pupae are found in the soil, which again makes them less amenable for control measures. The larvae nearing pupation have a habit of jumping and wriggling and move away from the surrounding host plant. As a result, the pupae are very much scattered in their distribution and escape from management practices. Moreover the adult flies spend most of their life span on non-host tree foliage and visit the field mainly for oviposition. Thus, the fruit flies have several adaptive factors favouring their survival and infestation potential. So biorational strategies that modify the fruit fly behaviour are the possible alternative methods of fruit fly management and they are also eco-friendly in nature.

Behavioural control using Trapping Technique

Trapping systems for insects are the important components in integrated pest management programmes. With the availability of sufficiently effective traps that capture both female and male fruit flies, trapping techniques may be used as behaviour control measures. Adults of tephritid fruit flies use chemical uses for finding the mate and use both visual and chemical uses to locate the host for feeding and oviposition. This phenomenon can be exploited for the trapping techniques against fruit flies.

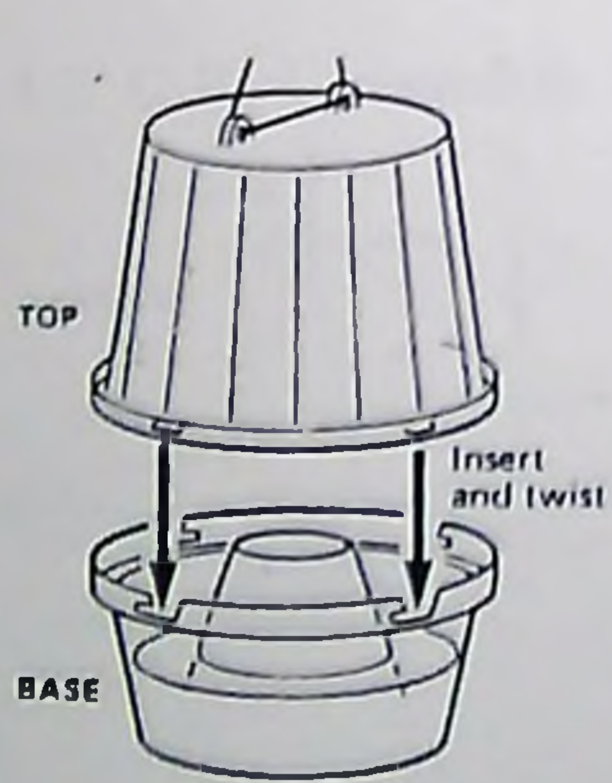
1. FRUIT FLY TRAPS

1.1Steiner trap



The trap was designed by Steiner (1952). It consists of a horizontal cylinder having a large openings at each end. The cylinder is slightly tapered so that both ends can be folded together for easy transportation. A cotton wick containing lure is suspended almost from the middle of the cylinder. Since, it has a large area for evaporation and large open area at each end, it helps in wider distribution of attractant vapour. The trap can catch a large number of flies in a short period of time.

1.2 McPhail trap

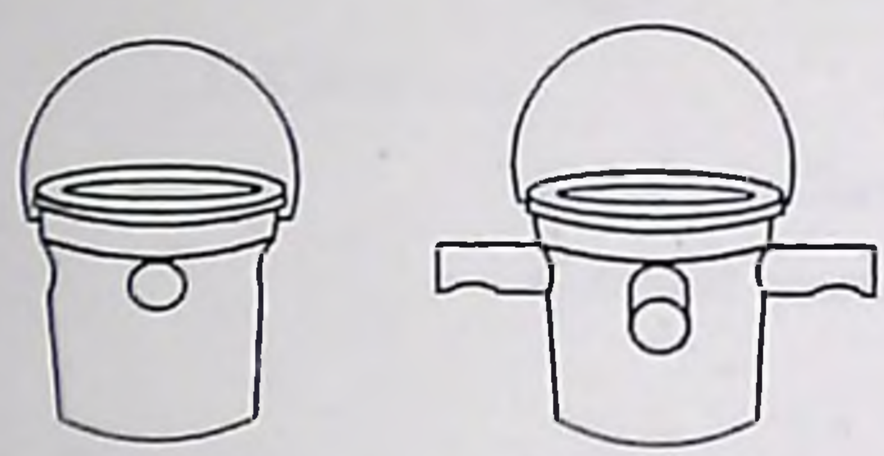


It consists of a medium sized glass or plastic flask with an invaginated entrance at the base. The lure mixed in water is placed at the bottom around the invaginated entrance at the base. Proteins hydrolysate is the common food lure used in this trap. The lure may be poisoned with pesticides like malathion. McPhail trap has been found useful in attracting a large number of flies but the disadvantage was that, the flies get immersed in the liquid and killed as compared to the Steiner trap, which can trap living specimens.

1.3 Jackson trap

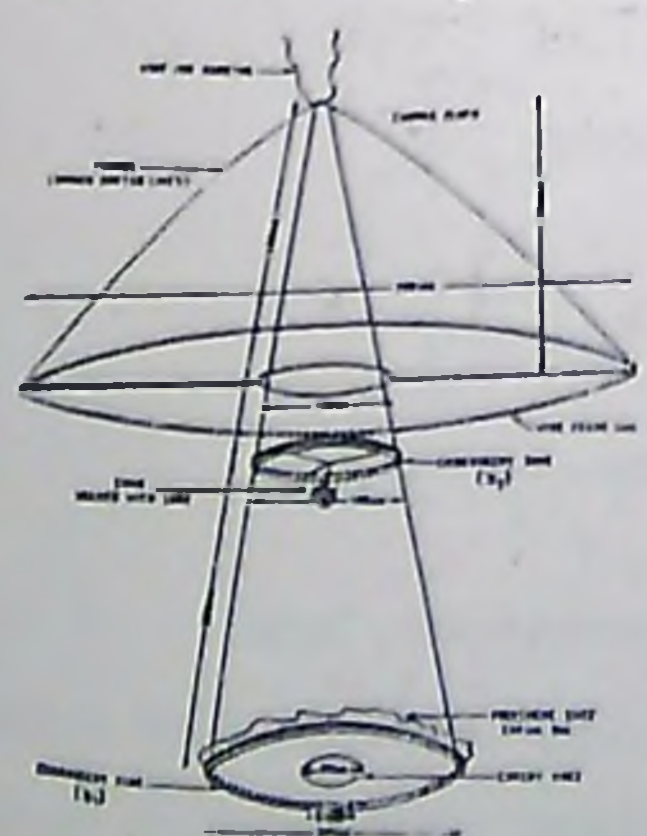
It is triangular or delta-shaped trap with sticky inner surface and fitted with a lure material. The inner surface is lined with a non-drying adhesive, which can trap the flies when they come close to the attractant. The outer surface of the trap may be coloured yellow or other attractive colours to increase the efficiency of traps. These traps are easy to assemble, cheap and are easily transportable. They are good for detection purposes especially for quarantine around airports and towns. Jackson trap is mostly used against Mediterranean fruit fly, *Ceratitus capitata*.

1.4 Bucket trap



These are simple, inexpensive traps of 1 to 2 liter capacity. They have 4 entrance holes, a wire handle and lure soaked cotton wick hung from the top. The flies attracted towards the lure material enters through the hole and killed by the pesticide and collected at the bottom of the trap. This trap can be modified accordingly to suit our needs.

1.5 G.K. Collapsible trap (Grewal and Kapoor)



It is a modification of Steiner trap and costs less than Rs.30 and is easy to transport. It consists of (a) conical bag of muslin cloth (750 mm long), (b) two embroidery rings of different diameters (300 and 180 mm), (c) wire with hook for swab to be fitted on the inner part of a smaller embroidery ring, (d) a circular (400 mm dia) transparent polythene sheet with a entrance hole (75 mm dia).

In dry weather the trap can be used as such, while in rainy season, a funnel made of polythene can be attached at the top of the trap to avoid wetting of the trap. The main advantage of this trap is the bag being of net cloth, there is a greater scope for the lure vapour to disperse and cover a larger area in shorter time. The trap can be folded and easily transported.

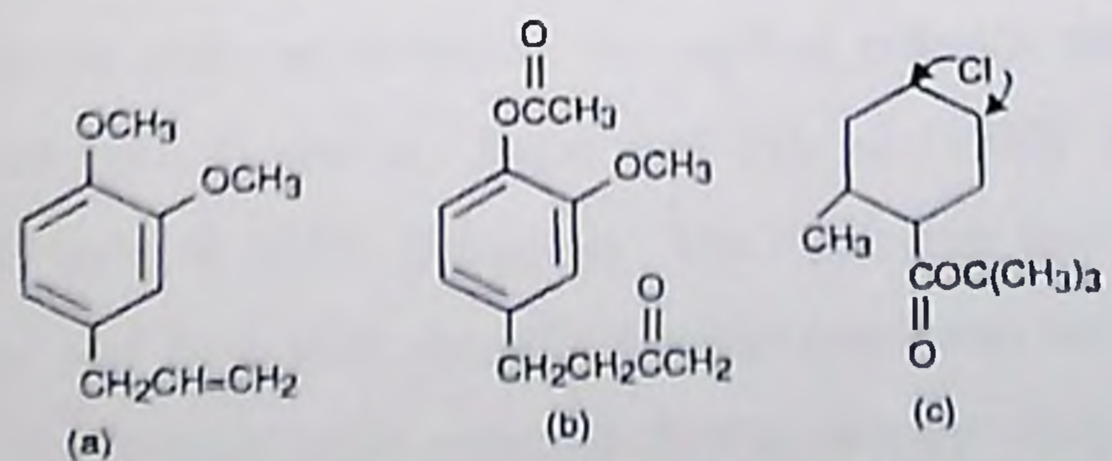
2. FRUIT FLY TRAPPING MATERIALS

Trapping materials used for fruit fly attraction can be categorised into

1) Parapheromone lures
   - Methyl eugenol
   - Cue-lure
   - Trimedlure
2) Food baits
   - Protein hydrolysate
   - Fruit pulp
   - Sugar
3) Visual uses
   - Coloured spheres
   - Coloured boards

2.1 Parapheromone lures

Parapheromones are chemicals, which cause responses similar to true pheromones, but actually they are not produced by the responding species.



Male lures for tephritid fruit flies : (*a*) methyl eugenol; (*b*) cue-lure; (*c*) trimedlure

These parapheromones attract only male flies of the responding species and widely used for male annihilation. Cue-lure, methyl eugenol and trimedlure are the commercially available parapheromones for the melon fly, *Bactrocera cucurbitae*, oriental fruit fly, *B. dorsalis* and Mediterranean fruit fly, *Ceratitis capitata* respectively. The process of reducing male population of fruit flies using parapheromone is often called Male Annihilation Technique (MAT), which involves the use of bait stations consisting of a male lure combined with an insecticide (usually malathion and more recently fipronil), to reduce the male population of fruit flies to such a low level that mating does not occur.

Hardy (1979) estimated that at least 90% of the fruit fly species are strongly attracted to either Methyl Eugenol or to cue-lure. Males of at least 176 species of fruit fly are attracted to Cue-lure and 58 species to methyl eugenol (Metcalf, 1990). Methyl eugenol is extremely effective as an attractant for male oriental fruit flies. These flies have been lured to traps from 0.8 to 1.6 km upwind (Steiner *et al.*, 1965). The oriental fruit fly, *B. dorsalis* was completely eradicated from the Commonwealth of Northern Mariana Islands in the year 1965. Fiberboard squares impregnated with 25 g of 97% methyl eugenol and 3% naled were distributed using aircraft at the rate of 115/sq. mile. The fiberboards were distributed 15 times at 2 weeks interval and resulted in the complete eradication of the species.

Outstanding success using this method have been recorded from places like California (Cunningham and Suda, 1986) and the Amami Islands of Japan (Iwahashi *et al.*, 1989). In the Pacific Island countries and Territories, eradication programmes were carried out against oriental fruit flies using impregnated coconut husk blocks. Coconut husk blocks of size 50 mm x 50 mm x 10 mm were impregnated with methyl eugenol and malathion and distributed from the air by helicopter at the rate of 400 per km^2, six times in 1999 and complete eradication was achieved (Allword, 2000).

Apart from male annihilation, the methyl eugenol traps were also used for studying the population dynamics. Bagle and Prasad (1983) studied the population dynamics of *B. dorsalis* in IIHR, Bangalore. The maximum population occurred during March, April, May and June with monthly average catch/trap being 1268, 270, 487 and 416 respectively. The lowest catch occurred during January, August and December with the catch of 42, 71 and 72 flies respectively. Further, the weekly catch of the fruit fly showed significant positive correlation with maximum temperature and negative correlation with relative humidity and rainfall.

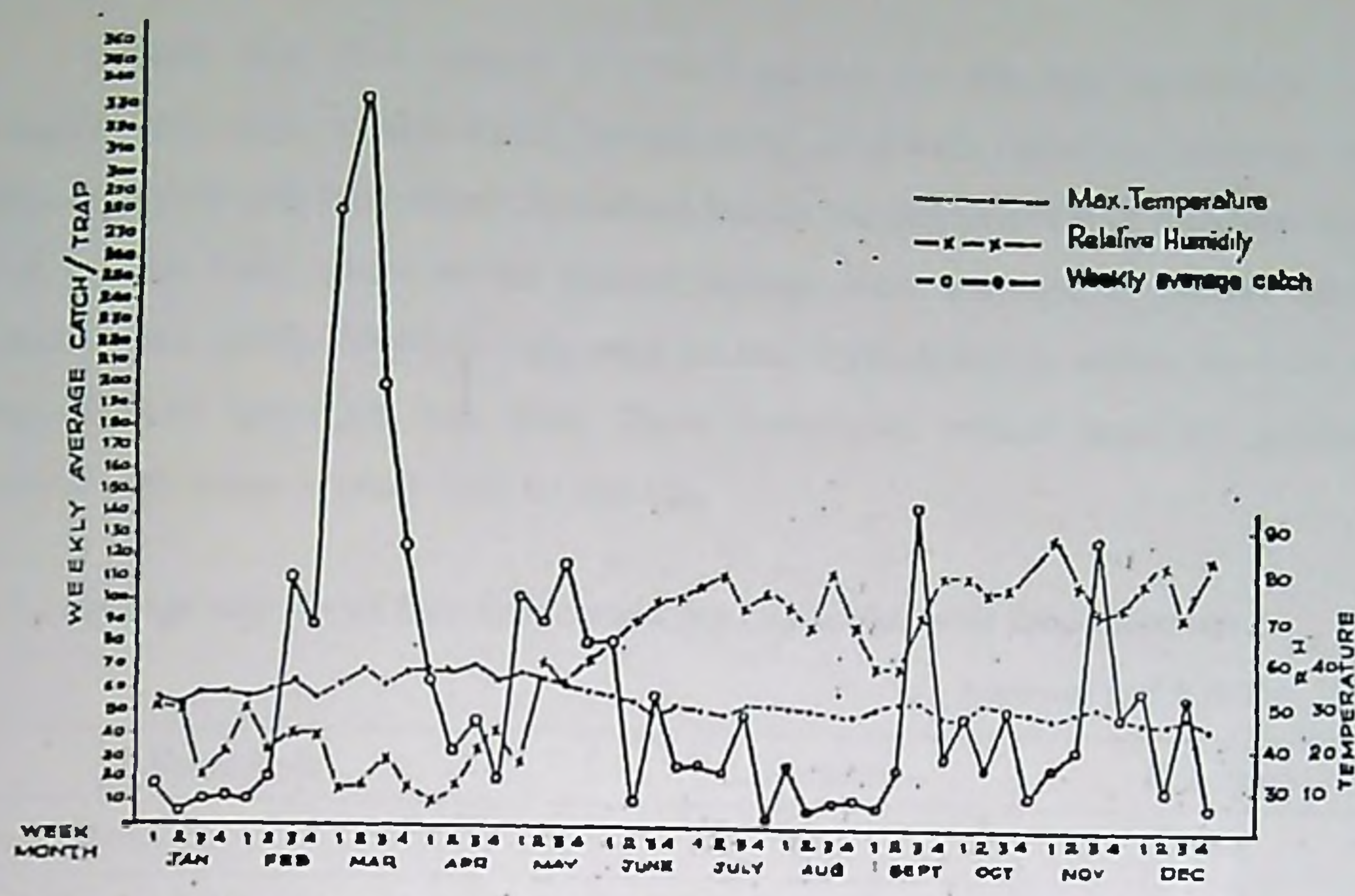


Fig. 1. Seasonal population dynamics of the Oriental fruit fly, *D. dorsalis* Hendel.

Miller *et al.*,(1993) found that ME produced carcinogenic effect in the liver of laboratory reared mice. A saturated analogue 3,4-dimethoxypropylbenzene was developed as substitute lure to the ME and possessed identical attractant properties (Mitchell *et al.*,1995). Reghunath and Indira (2000) reported methyl eugenol analogue from the holybasil (*Ocimum sanctum*) 20 g of crushed leaves with 0.5 g each of citric acid and of carbofuran 3 g in 100 ml of water at four traps/ha was found to be attractive to *B. dorsalis*.

Vargas *et al.* (2000) studied the *B. cucurbitae* capture using bucket traps baited with Cue-lure. Wicks with Cue-lure and malathion appeared hard and unattractive after 8 weeks of weathering in field. The weekly catch of *B. cucurbitae* varies from 543.3 to 803.8 per trap. Pawar *et al.* (1991) reported that, the sex attractant trap Cue-lure was more effective in trapping melon flies than the food attractant trap tephilure and the fruit fly catch depends on maximum temperature, day humidity and rainfall.

Jackson traps baited with trimedlure found to be effective in trapping males of Mediterranean fruit fly, *Ceratits capitata* and the per day catch was as high as 78 males per trap (Liquido *et al.*, 1993).

2.2 Food baits

Female fruit flies require a protein source for the egg production. The physiological state (age, mating status, hunger level) of female tephritids influence their attraction to protein and fruit odours. Immature female Mediterranean fruit flies were more attracted to host fruit, where as the mature females were attracted to protein odours (Cornetius *et al.*, 2000). McPhail traps with protein hydrolysate is widely used for the trapping of most temphitid fruit flies. These hydrolysed protein baits are generally attractive to both sexes of many fruit fly species.

Table 1 . Average number of fruit flies caught per trap baited with food materials

Agarwal and Kumar (1999)

Food bait	*B. dorsalis*	*B. zonata*
Protein hydrolysate	44	148
Fermented palm juice	9	76
Sugar	10	16
Ripe mango pulp	25	132

However, there are certain disadvantages in the use of protein hydrolysate. Degradation of protein source and effectiveness over a short distance are the main drawbacks (Heath *et al.*, 1995). Trapped fruit flies are offers found severely decomposed, increasing difficulty in species identification and removal of trapped insects require considerable effort. Protein baits also attract a number of non-target insects and considerable time is required for sorting of insects.

Heath *et al.* (1995) evaluated dry trap baited with two-component food-based synthetic attractant for Mediterranean and Mexican fruit flies. The two-component mixture containing blend of ammonium acetate and putrescine was more attractive than McPhail traps with standard protein bait.

A four-component synthetic attractant made of ammonium bicarbonate, linoleic acid, putrescine and pyrrolidine was developed by Wakabayashi and Cunningham (1997). It was found to be attractive for both sexes of melon fly and was as effective as the standard food bait.

The recent trend in trapping techniques of fruit flies is the female targetted trapping systems. Epsky *et al.* (1999) conducted studies on field evaluation of female targetted trapping system for Mediterranean fruit fly *Certatitis capitata* in seven countries (Greese, Honduras, Mauritius, Morocco, Portugal, Spain and Turkey). The food lure used was a three-component attractant consisting ammonium acetate, putrescine and trimethylamine. Traps baited with the three-component attractant captured equal or greater numbers of females than McPhail trap baited with protein hydrolysate and the females accounted for more than 90% in the total capture.

2.3 Visual traps

Adults of fruit flies use both visual and chemical uses for locating the host for feeding and oviposition. Fruit mimicking colour spheres coated with sticky materials are useful devices in attracting and trapping the fruit flies (Prokopy, 1968).

Table.2 Attractive fruit mimicking colour spheres

Colour	Fruit fly	Reference
Dark red	Apple maggot fly *Rhagoletis pomonella*	Prokopy, 1968
Green	Walnut husk fly *Rhagoletis completa*	Riedl *et al.*,1985
Red & yellow	Olive fly *Dacus oleae*	Prokopy and Haniotakis, 1975
Yellow & black	Med fly *Ceratitis capitata*	Katsoyannos, 1989
Yellow	Mexican fruit fly *Anastrepa ludens*	Cytryowicz *et al.*, 1982

Oriental fruit fly, *B. dorsalis* captures were higher on yellow and white spheres coated with Tangle foot than orange, red, blue and black ones hung in guava orchard. Captures were greater on 4 cm yellow spheres than that of 2 cm (Vargas *et al.*, 2000).

Table.3 Mean number of *B. dorsalis* captured on colour spheres

Sphere colour	No. of *B. dorsalis*/sphere/tree x ± SEM
Yellow	11.9 ± 3.9
Blue	5.0 ± 2.0
Green	3.0 ± 2.6
Orange	2.0 ± 1.0
Red	1.0 ± 0.3

Duan and Prokopy (1995) developed pesticide treated colour spheres for the control of Apple maggot flies *R. pomonella*. Red spheres treated with a mixture containing 1% methomyl, 59% corn syrup and 40% latex paint effectively trapped the maggot flies and the effect persisted for 35 days. However the spheres were ineffective after exposure to 6.6 mm rainfall.

Increasing trapping efficiency by combination of lures

Fruit flies often use more than one type of cue for host identification. Hence by combining more than one type of cues, for example visual and chemical cues, the efficacy of trapping system could be considerably increased.

The response of male oriental fruit fly *B. dorsalis* to coloured bucket traps baited with methyl eugenol was determined in guava orchard (Stark and Vargas, 1992). Plastic bucket traps were painted with white, black, yellow, orange, blue, green and red colours. Wicko containing 1 ml of methyl eugenol mixed with 0.5% nalad were placed in each trap and flies were collected every 24 h for 8 days. White and yellow traps caught the largest number of flies while green, red and black caught the fewest.

Liburd *et al.* (1998) combined colour sticky traps with ammonia for increasing the trapping efficiency against Blue berry maggot, *Rhagoletis mendax*. Polycon dispensor containing 5.6 g of ammonium carbonate baited along with Pherocon Yellow Board, Green, red and yellow spheres were evaluated against the coloured traps alone. Significantly more flies were captured on ammonium carbonate baited colour spheres than the unbained ones. Among the baited spheres, red spheres baited with ammonium source captured more flies, which is followed by green and yellow spheres.

DISCUSSION

Q 1. Give details regarding the important species of fruit flies with special reference to Kerala?

So far 320 species of fruit flies were known from various parts of India. In Kerala, the important fruit flies were melon fly, *Bactrocera cucurbitae*, oriental fruit fly, *B. dorsalis*, guava fruit fly, *B. zonata* and *Dacus ciliatus*.

Q 2. Since, the male lure, methyl eugenol is carcinogenic, how does it become the component of environmentally safe behavioural control?

The methyl eugenol is not directly sprayed on the fruit on vegetable crops. It is safely kept inside the trap. But, while handling the trap, one should be very careful. This is a caution notice only and there will not be any direct health risk due to methyl eugenol kept inside the trap.

Q 3. Does the colour of the visual cure vary in different crops for the same fruit fly?

It may vary according to the crop and location. A thorough behavioural study should be undertaken for each fruit fly under different crop situations before recommending a best visual cure trap.

Q 4. What about parasitoids and predators for fruit fly?

There are reports of parasitoids like *Diachasmimorpha longicaudata, Pcytallia fletchesis* for fruit flies. But these are occurring in natural condition and there is no report on mass culturing and release of parasitoids for the control of fruit flies.

Q 5. Whether the tulsi leaf-extract can be used for the control of all fruit flies?

No, tulsi leaf contains only the methyl eugenol analogue only. So, the fruit flies that are attracted to methyl eugenol (like *B. dorsalis*) can be controlled using tulsi leaf extract.

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COCCINELLID PREDATORS AS POTENTIAL BIOCONTROL AGENTS

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SEMINAR REPORT

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ABSTRACT

Pest management strategy was given a boost with the extensive practice of biological control, where predators, parasites and pathogens play a major role in regulating the pest population. Predatory coccinellids commonly called as ladybird beetles are linked to biological control more than any other taxa of predatory organisms. Coccinellids are important natural enemies of pest species, especially whiteflies, aphids, mealy bugs, scales and mites. The first spectacular success in biological control was by the introduction of *Rodolia cardinalis* against cottony cushion scale in citrus in 1887-'88.

Coccinellids can be effectively utilized in biocontrol programmes in three ways, viz., natural biological control, conservation and augmentative biological control. In nature, management of various sucking pests is effected by coccinellids in many agro-ecosystems without any human intervention. Conservation of coccinellids is achieved by reducing the use of toxic pesticides, selective use of resistant varieties and suitably modifying the cultural practices. Importation and periodic release are the two methods in augmentative biocontrol. It involves mass multiplication and release of coccinellids. Several coccinellids are successfully reared in laboratory conditions on living hosts or on artificial media. *Cryptolaemus montrouzieri*, which is widely released in orchards for the management of mealy bugs and scales is mass multiplied in the laboratory on the mealy bug, *Planococcus citri* on pumpkins.

In our crop fields a more practicable method of biological control is conservation, which augurs well with other ecofriendly management measures. This involves selective use of management tractics which enhance the population and also efficiency of coccinellid predators in the field. Augmentative biocontrol require improvement of mass production methods of coccinellids.

COCCINELLID PREDATORS AS POTENTIAL BIOCONTROL AGENTS

INTRODUCTION

In this era of pest management where environment friendly management practices are more relevant in agriculture, biological control gains momentum among many other tools in IPM. In biological control parasites, predators and pathogens play a major role in regulating the pest population without any adverse effect to the ecosystem. Among the predators in nature, members of the family coccinellidae ranks first in their predatory efficiency.

They are brightly coloured in various shades of red, black and brown and are usually spotted. Hence they are commonly called as lady bird beetles. Both the grubs and adults are voracious feeders on various sucking pests like aphids, mealy bugs, scales etc. Though they exhibit diverse feeding habits majority of them are carnivorous and in nature many phytophagous insects are kept partly or completely under check in crop plants without any human intervention. The classical example of biological control using a predator involves the use of coccinellid beetle, *Rodolia cardinalis* to control cottony cushion scale.

Historical perspective

The value of lady bird beetles as natural predators has been recognised since mid 19th century. The first well planned and successful biological control attempt was undertaken in 1887-88, when citrus industry in California was seriously threatened by cottony cushion scale. The Australian ladybird beetle, *Rodolia cardinalis* was introduced into California within a year spectacular control was achieved .

In India, the earliest attempt at introduction of a natural enemy was in 1898 when a coccinellid, *Cryptolaemus montrouzieri* from Australia was released against coffee green scale, *Coccus viridis*. It is now a common mealy bug predator in South India. The same *Rodolia cardinalis* was introduced in India against cottony cushion scale against the same cottony cushion scale on wattle of commerce in Nilgiris.

The establishment of regional station of Commonwealth Institute of Biological Control at Bangalore in 1957 gave an incentive to biocontrol work in India.

The beneficial role of parasites and predators in regulating pest populations and practical methods using them in regulating pest populations and applying them in the field, were investigated. This work got a further boost with the establishment of All India Co-ordinated Research Project on Biological Control of crop pests and weeds in 1977. The AICRPBC was further upgraded to the level of Project Directorate of Biological Control in 1993.

PDBC- Co-ordination centres

1. IIHR Bangalore
2. CPCRI Kayamkulam
3. SBI Coimbatore
4. CTRL, Rajamundri
5. IISR, Lucknow
6. IARI, NewDelhi
7. PAU, Ludhiana
8. GAU, Anand
9. KAU, Trichur
10. APAU, Hyderabad
11. TNAU, Coimbatore
12. MPAU, Pune
13. GBPAU&T, Pantnagar
14. Aau, Jorhat

Like any other biological control agent coccinellids can be effectively used in 3 days.

1) Natural Biological Control
2) Conservation
3) Augmentative Biocontrol

1) Natural Biological Control

In nature even without the intervention of man, regulation of pest population occurs. The pest population is maintained within certain upper and lower limits by the action of abiotic and biotic factors. This is referred to as natural control.

In our crop fields coccinellids play a vital role in the natural control of various pest species especially aphids, mealy bugs and scales. Research conducted in various ecosystems revealed that coccinellid beetles effectively control major sucking pests in nature. The following reports highlight their role in the natural control of crop pests.

Rice

Several species of coccinellids were reported in rice effectively predating naturally on a number of species of pests in rice.

Coccinella arcuata F. a predator of rice plant hoppers *Sogatella furcifera* and *Nilaparvatha lugens* was recorded in 1966 and 1967 at Cuttack. In 1973 Abraham *et al.* recorded *Coccinella arcuata* a predator of BPH for the first time in Kerala. Mammen and Nair (1977) reported that the adults of *C. arcuata* as an efficient predator of nymphs adults of *Baliothrips biformis* on rice in Kuttanad tract, Kerala. This is the first record of coccinellidae predating on the thrips.

Coccinella rependa and *Menochilus sexmaculatus* were predacious on *Nilaparvatha lugens* in Mandya (Manjunath, 1979). Adult of *Brumoides suturalis* were found preying on nymphs and audlts of *Sogatella furcifera* and nymphs of *Nephotettix virescens* in New Delhi. *Micraspis discolor* preferred third instar nymphs of the Delphacid as prey and consumed and average of 47 of them during entire larval stage under laboratory condition.

Brumoides suturalis, Coccinella septempunctata, Menochilus sexmaculata and *Scymnus* sp. were found associated with leaf and plant hoppers(Kaushik *et al.*, 1986).

Micraspis sp. has been reported as the most important coccinellid predator in different rice ecosystems of Kerala (Ambikadevi *et al.*, 1998, Bhaskar, 1999, Beevi *et al.*, 2000a).

Studies on RRS, Moncombu and farmer's fields of Kuttanad during khariff and rabi seasons of 1996 and 1997 revealed the presence of large number of natural enemies of rice pests (Ambikadevi, 1998). Among the natural enemies coccinellid predators, *Micraspis* sp. and *Menochilus sexmaculatus* showed higher rates of predation.

In a survey undertaken in Thrissur district to quantify the abundance, diversity and relative occurrence of pests and natural enemies in different rice agro-ecosystem (Beevi *et al.*, 2000a) spiders, coccinellids and mirid bugs constituted the major portion of predators.

Cowpea

A survey conducted at College of Agriculture, Vellayani recorded that six predators were reported to be associated with pea aphid, *Aphis craccivora*, on cowpea. Four among the six predators were coccinellids. They were *Cheilomenes sexmaculatus*, *Coccinella septempunctata*, *Scymnus* sp. and *Micraspis crocea*. The predator *Cheilomenes sexmaculatus* showed better field establishment and performance due to the nature of interaction with ecosystem which conferred advantageous.

Population counts of pests and natural enemies were taken from insecticide sprayed plots and also from unsprayed plots. Predators associated with *Aphis craccivora* were *Menochilus sexmaculatus* and *Micraspis crocea*. Incidence of pest was more in unsprayed plots but the population of the pest remained higher only for a short period and it was effectively checked by natural enemies, the population of which synchronized with that of the pest.

Chilly

Survey conducted at Instructional Farm, College of Agriculture, Vellayani and neighbouring farmer's fields revealed that spiders and coccinellids (*Menochilus sexmaculatus* and *Micraspis crocea*) were the major natural enemies associated with the chilly agro-ecosystem.

Banana

Seven species of coccinellids were found in aphid colonies. They are *Scymnus quadrillum*, *Menochilus sexmaculatus*, *Adalia bipunctata*, *Coccinella septempunctata*, *S. pyrocheilus* and *Nephus luteus* (Padmalatha, 1998).

Coconut

Black beetle, *Chilocorus nigrita* was effective to control coconut scale, *Aspidiotus destructor* (Pandian *et al.* ,2002)

Cotton

In cotton ecosystem *Menochilus sexmaculatus* and *Coccinella septempunctata* were reported as important natural enemies in aphid colonies (Yadav, 1999).

Sugarcane

In sugarcane aphids were minor pests probably due to the activity of predators (Lim and Pau, 1978). Coccinellids are reported to be the important mortality factors.

Orchard systems

Successful biocontrol programmes were done in orchard systems. Many sucking pests were kept at a lower level by the predatory coccinellids. Bhaskar (2001) reported that several coccinellids from guava and sapota orchards. *Cryptolaemus montrouzieri* is the major predators in these ecosystems.

Cheilomenes sexmaculatus, Jauravia soror, Chilocorus nigrita and *Scymnus nubilis* were associated with several pests like *Aphis gossypii, Coccus viridis, Icerya purchase, Ferrisia virgata* and *Chloropulvinaria psidii*. In sapota orchards *Coccus viridis* and *Chloropulvinaria psidii* were predated by *Cryptolaemus montrouzieri* and *Chiloconus nigrita* (Bhaskar *et al.*, 2001).

For apple wooly aphid *Coccinella septempunctata* and *Hippodamia variegata* are found to be the major predators (Mani and Krishnamoorthy, 2002).

Redgram

Bhaskar and Viraktamath (2000) reported coccinellids from redgram ecosystem they are *Rodolia fumida* on scale, *Brumoides suturalis* on aphid, *Aphis craccivora* and *Cheilomenes sexmaculatus* on mealybug.

2) Conservation

Conservation of natural enemies involves environmental modification to benefit natural enemies. Minimal disruption is attained by

(a) reduction of impact of pesticides

(b) Selective use of pest resistant varieties

(c) Changes in cultural practices including the maintenance of refugia for natural enemies through the use of strip planting, field borders or cover crops.

(d) Alteration of regional landscapes.

Pesticides

Among the cultural practices pesticide application has the greatest effect on local population of coccinellids. So the greatest gains may be attained through reduction of toxic pesticides in the habitats. Insecticides and fungicides reduce the coccinellid populations. They may have direct or indirect toxic effects. Surviving coccinellids may also be directly affected, eg. reduction in fecundity, or longevity or indirectly affected by decimation of their food sources. Adults may disperse from the treated areas in response to severe prey reductions, or because of insecticide repellency.

Coccinellids rapidly recolonize agricultural fields after insecticide applications if sufficient preys are present. Although often less susceptible to insecticides than are their prey, predacious coccinellids are highly susceptible to several insecticides applied to cotton, citrus and fruit orchards. Insecticide selection and application timing are used to minimize effect on coccinellids.

Thomas and Phadke (1996) reported that Quinalphos was less toxic to *Coccinella septempunctata* than chlorpyriphos and oxydemeton methyl.

Shukla *et al.* (1994) reported that recommended field dosages of the most commonly used insecticides like monocrotophos, quinalphos and methyl parathion caused mortality to adults of the predator *Coccinella septempunctata*. Malathion and endosulfan was harmful to the predators but to a lesser extend.

Singh and Saxena (1997) studied side effects of neem products on insect pathogens and natural enemies of spider mites and insects. Based on the study they found that under field conditions side effects on nymphs or larva are usually slight or even not significant.

In a study conducted at UAS, Bangalore (Diraviam, 1999) reported that *Beauveria bassiana* was harmful to *Curinus coeruleus*.

Pest resistant varieties

Prey reared on resistant plants may have significant impact on the fitness of coccinellids. Plants may directly or indirectly influence coccinellid effectiveness as a result of altered prey suitability or host finding success of the predator. Searching behaviour is influenced by the complexity of the substrate searched. Predation of aphids was reduced on plants with dense hairs and trichomes. But reduction varies with coccinellid species and developmental stage. Obrycki (1986) conducted laboratory, greenhouse and field studies to assess the interaction between the potato hybrids bearing various densities of glandular trichomes and natural enemies. Under green house conditions mobility of newly hatched coccinellid larvae were inversely related to the density of glandular trichomes. The same coccinellid species (*Coccinella septempunctata*) oviposited on pubescent clones in the field conditions. Bhaskar (1999) reported that *Micraspis discolor* is feeding more BPH on the resistant rice cultivar, Kanakom than on other varieties like Jaya and Jyothi.

Cultural practices

Changes in cultural practices including the maintenance of refugia for natural enemies through the use of strip plantings, field borders or cover crops. Establishment of refuges during a production season may provide increased pest suppression by coccinellids. Strips of weeds within crops may harbor significant populations of alternate prey and beneficial insects, this practice reduced pest aphid population in apple (Wyss, 1995).

Reduction of *Lygus* sp. in cotton in early spring using weed management has been proposed because predators are emigrating when pest densities are increasing. Selection of ground cover under an arboreal crop may or may not influence coccinellid population in the crop. Coccinellids may be abundant in refuges at various times of the year, particularly where large number of over wintering adults is common. However, coccinellids may occur in numerous habitats seasonally, and refuge may be considered as any area other than the focus crop or field. Careful attention of over wintering sites and banded herbicide applications conserve the predator populations in apples without enhancing refugia for the pest.

Cropping pattern

The broad host range and high mobility of adult coccinellids, make it difficult to partition the mortality caused by coccinellids in any given ecosystem. Only evaluation of the population dynamics of a target coccinellid species at the landscape level will provide reliable predictions of coccinellid impacts in any target system. In central Mexico two *Hippodamia* species were more prevalent in maize-faba bean polycultures than in corn monocultures, presumably because of the availability of extra floral nectaries in faba beans (Trujillo-Arriega, 1990).

3. Augmentation

Augmentative biological control involves introduction of exotic biological control agents or production and utilization of indigenous biocontrol agents. It is defined as the efforts to increase the populations of natural enemies either by propagation and release or by environmental manipulation. These effects can be achieved by releasing additional numbers of a natural enemy into a system or modifying the environment in such a way to promote effectiveness. Releases have to be done periodically.

Importation

Importation is the introduction of exotic natural enemies into an area either to control an exotic pest or an indigenous pest. Successful biological control of the cottony cushion scale by *Rodolia cardinalis* gave more importance to importation programmes focused on coccinellids. *Cryptolaemus montrouzieri* was introduced from Australia to control mealy bugs in fruit crops like citrus, grapes, guava etc. For the control of *Melanaspis glomerata* in sugarcane *Chilocorus nigritus* was introduced and *Curinus coeruleus* for managing the subabul psyllid, *Heteropsylla cubana*.

Prerequisites for importation

- Field observation of predation on the target pest and consistent association of coccinellid and prey species in their native range.
- Alternate food materials like non-target insect prey, microorganisms associated with certain prey, fungi pollen and nectar may be required by an introduced coccinellid species.
- Releasing climatically matched species or biotypes, including those adapted to local temperature conditions has been considered critical for biological control.
- Coccinellids feed more than one prey species, thus disruption of existing biological control by introduced coccinellid species and potential for indigenous coccinellid species to disrupt the introductions needs to be considered.
- Several species of parasitic Hymenoptera, Diptera, Nematoda and Acarina attack coccinellids.

Periodic release

Periodic release is the mass multiplication and release of biocontrol agents. It may be either inoculative or inundative. We have to use potential predators for periodic release. Predatory potential of some important coccinellids are given below.

Cryptolaemus montrouzieri (grubs) - 900-1500 mealy bug eggs or 300 nymphs or 30 adults

Chilocorus nigritus (adult) - 125 scales/day

(grubs) - 46 scales/day

Coccinella septempunctata (adult) - 41 aphids/day

Cheilomenes sexmaculatus - 33 aphids/day

Mass multiplication techniques

There are two ways for mass multiplication of coccinellids.

1. *In vivo* - Rearing the coccinellids on living host or the prey.

2. In artificial media - mass rearing is done in different media. The trials are still going on. Till date not much success is achieved.

Prerequisites for selection of a biocontrol agent

- Should be the effective species or strain amenable for mass production
- Capable of developing even on resistant/tolerant cultivars
- Compatible with other crop production inputs
- High reproductive potential
- Suitable for particular climate niche
- Preferably tolerant to most common insecticides
- Suitable for seasonal innoculative/inundative use

Mass production of *Cryptolaemus montrouzieri*

Cryptolaemus montrouzieri

Cryptolaemus is called as mealy bug destroyer. Both adults and grubs are predating all the stages of mealy bug. Grubs are voracious feeders. Grub consumes 900 to 1500 mealy bug eggs, 300 nymphs or 30 adults in its lifespan. It is native to Australia. In 1892 introduced to California to control citrus mealy bugs. Then it was introduced to India. Mass culture is a prerequisite to release the predator in large numbers.

(a) Mass culture of Host Insect (Mealy bug)

Mealy bugs can be produced in large numbers on potato sprouts or ripe pumpkins *Planococcus citri* is adopted in insectary programmes than other mealy bug species. Pumpkins are selected with ridges and grooves with a small stalk, which makes handling easy. The pumpkins are dried under shade. Each pumpkin is infested by with *P. citri* crawlers maintained infestation room. Each of the infested pumpkin is placed in 30 cm^3 wooden cage, all sides covered with white cloth or white nylon mesh. They get covered with mealy bugs in 15 days.

(b) Production procedures of predator

After 15 days of infestation of pumpkin with mealy bug, they are exposed to a set of 100 beetles for 24 hours. After exposing the pumpkin is kept back in the cage. The beetle during the period of exposure feed on mealy bugs as well as deposit their eggs singly or in groups of 4-12. The grubs after emergence feed on eggs and small mealy bugs but as they grow they become various, and feed on all the stages of mealy bugs. For facilitating pupation, provide dried guava leaves or pieces of papers at the base of each of the cages. The first beetle start emerging on 30th day of exposure to *C. montrouzieri* adults. The beetles are collected daily and kept in separate cages for about 10-15 days to facilitate completion of mating and oviposition.

The beetles are also feed with a diet containing agar (1 g), sugar 20 g, honey 40 cc and water (100 cc). The adult beetle diet is prepared by boiling water in 70 cc of water, adding 1 g agar, diluting 40 cc honey in 30 cc of water and adding to the sugar and agar mixture when it comes to boiling point. The hot liquid diet is kept on small white plastic cards in the form of droplets, which get solidified on cooling. Such cards can be fed not only to C. montrouzieri but to many other species of coccinellids from each case 175 beetles are obtained. Emergence of beetle is completed in 10 days. The cost of production of 100 beetles at 1998 price index was Rs.10.

Beetles can be reared on *Corcyra cephalonica* egg but empty ovisacs of *P. citri* are kept for inducing egg laying by the beetles. The beetles are also multiplied on semi synthetic diet which is still in the process of further refinement.

Precautions

1. Avoid scarcity of food for the grubs to avoid cannibalism by grubs.

2) All the pumpkins showing signs of rotting should be properly incinerated.

Field release

Before releasing in field in the endemic areas, moderate to severely infested plants are marked. Plant trunks are ringed one foot away with a band of insecticidal dust 24 hours before the release of beetles. This stops patrolling of ants on the trunk at least for 3 days.

Release

Adult release is practiced for cocccinellids.

Precautions

Follow the precise manner, time and frequency of application. Releases should be done in early morning or in evening times. In situations where pesticides have been applied for containing out breaks, bioagents could be utilized after a safe weighting period.

The recommended biological control systems

Crop / pest	Biocontrol agent	Dosage	Frequency
Coffee			
Mealy bug, *Planococcus* and *Pseudococcus* sp.	*Cryptolaemus montrouzieri*	2-10 adults/infested plant	After the blossom
Arecanut			
Ischnaspis longirostris	*Chilocorus nigrits*	20/50 adults/plant	Release as soon as infestation is notied
Citrus			
Planococcus citri *Iceryia purchase*	*Cryptolaemus montrouzieri* *Rodolia cardinalis*	10 beetles/infested plant	After the blossom
Grapes			
Maconellicocus hirsutus	*Cryptolaemus montrouzieri*	2500 to 3000 beetles/ha	As soon as infestation is noticed
Guava			
Chloropulvinaria psidii	*Cryptolaemus montrouzieri*	10-20 beetles/ infested plant	As soon as infestation is noticed
Subabul			
Heteropsylla cubana	*Curinus coeruleus*	20-25 adults/tree	Twice during July and October
Sugarcane			
Melanaspis glomerata	*Pharoscymnus horni* *Chilocorus nigrita* *Sticholotis madagasse*	1500 adult beetles/ grubs	As soon as infestation is noticed
Apple			
Quadraspidiotus perniciosi	*Chitocorus infernalis*	20 adults/50 grubs per tree	Once in April-May

Sources of supply of biocontrol agents

Private units

Government units and

NGOs

CONCLUSION

Coccinellids can be effectively utilized in natural biological control, conservation and augmentation. Natural biological control will continue to play a role in nature without any human intervention. Conservation can be achieved by reducing the use of toxic pesticides and changing the cultural practices. Augmentation is practiced whenever the pest population increases beyond our control. Coccinellids are the potential natural enemies which can be introduced into an area wherever a new Homopteran pest causes threat. They have the high potential for biological control and can be recommended for future programmes.

DISCUSSION

1. What are the major predatory coccinellids seen in Kerala?

In different ecosystems in Kerala we have a large number of coccinellids. In cowpea, *Cheilomenes sexmaculatus, Coccinella transversalis, Scymnus* sp., *Jauravia* sp. are the major predators for cowpea aphid. *Scymnus* sp. and *Jauravia* sp. are the predators in bittergourd. For BPH and other hoppers in rice field, *Micraspis crocea, Cheilomenes sexmaculatus* and *Coccinella transversalis* are the major ones.

2. Is there any report on insecticide resistance in coccinellids?

Coccinellids are resistant to botanicals and myco insecticides. They are less susceptible to the insecticides than their prey. But some commonly used insecticides cause high mortality to these predators.

3. What are the coccinellid predators of BPH in rice field?

Micraspis sp., *Cheilomenes sexmaculatus, Coccinella transversalis, Brumoides suturalis, Harmonia octomaculata* are the coccinellid predators in rice fields.

4. Which are the parasitoids attacking the coccinellids?

Several insects in the orders Hymenoptera, Diptera and Nematodes are attacking the coccinellids. We have to take care during importation to avoid the introduction of these parasitoids.

5. Which among the three methods of biocontrol can be practiced in Kerala?

Conservation is the method which is best suited to Kerala condition. In our crop fields lot of natural enemies are already present. By reducing the use of highly toxic pesticides and slightly modifying our cultural practices we can achieve that.

6. Whether augmentative biocontrol is needed in Kerala?

So far we have not done any augmentative programmes. For the coccinellids available in Kerala mass culturing is not standardized. We have to refine our technology to practice augmentation. Whenever the pest population exceeds beyond a level we can adopt this as a last measure.

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HYMENOPTERAN PARASITOIDS OF RICE PESTS - THEIR IMPORTANCE AND SUCCESS IN BIOLOGICAL CONTROL PROGRAMME

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ABSTRACT

Biological control involves the purposeful utilization of natural enemy components for the suppression of pest population. These natural enemies include predators, parasitoids and pathogens of which parasitoid play an important role in biological suppression of crop pests. The greatest diversity of parasitoids are seen in the Order Hymenoptera. In classical biological control programme about 393 species of parasitoids were listed of which 344 were found to be hymenopterans (Greathead, 1986).

A survey conducted at different locations of Thrissur District indicate the abundance and diversity of hymenopteran parasitoids in rice ecosystem (Beevi *et al.*, 2000). Trichogrammatids, platygasterids, scelionids, eulophids, etc. are the most common and important parasitoids seen in rice ecosystem. Experiments conducted with *Trichogramma japonicum* and *T. chilonis* against rice stem borers and leaf folders gave promising results (PDBC, 2000). Laboratory studies were conducted for the mass rearing of parasitoids such as *Platygaster* sp., *Tetrastichus* sp., *Anagrus* sp. etc., but the mass production and field release techniques are yet to be standardised.

The most important method of biocontrol is the conservation of existing natural enemy fauna in the rice ecosystem. This can be achieved by selective, rational and discriminative use of insecticides, which will enhance the population of these hymenopteran parasitoids in the field.

HYMENOPTERAN PARASITOIDS OF RICE PESTS - THEIR IMPORTANCE AND SUCCESS IN BIOLOGICAL CONTROL PROGRAMME

INTRODUCTION

Residue, resistance and resurgence are the major harmful effects due to indiscriminate use of pesticides. These reasons led entomologists to think and seek an alternate and more acceptable methods of which biological control is the major aspect.

Biological control involves the purposeful utilisation of natural enemy components so as to reduce the pest population. Natural enemies include parasitoids, predators and pathogens of which parasitoids have got an important role.

A parasitoid is an insect that parasitises other insects and arthropods. A parasitoid is parasitic in its immature stages, but the adults are mostly free living. They attack any host stage, but the adult stage is least frequently parasitised. They are most frequently used in biological control than any other kind of bioagents.

The greatest diversity of parasitoid is found in the order Hymenoptera. In classical biological control programme, 393 species of parasitoids were listed, of which 344 species are found to be hymenopterans (Greathead, 1986).

Successful Hymenopteran parasitoids utilised in classical biological control programme

Parasitoid	Pest
Eucarsia pernciosi *Aphytis* sp. nr. *proclia*	*Quadraspidiotus perniciosus*
Aphelinus mali	*Eriosoma lanigerum*

Characteristic features of Hymenopteran parasitoids

1) They are very minute wasps, generally having a size of 1 - 2 mm
2) Host specificity - Most of them have a narrow host range, only one or few hosts is required for the complete development of a parasitoid.
3) Females show very high host searching capacity.
4) They are adapted to varied terrestrial conditions.

5) They are active even at low host population level.
6) Hyper parasitism - Some of them parasitise the primary parasitoid of pests, there by reducing the parasitic potential of the primary parasitoids.

In all most all ecosystems, the population and diversity of these parasitoids are very high. Beevi *et al.* (2000) conducted a study to estimate the diversity of hymenopteran parasitoidsin rice ecosystem. The observations of the study are given in the table.

Hymenopteran diversity in rice ecosystem

Location	Hymenopterans at weekly intervals (Nos)				Total Nos.	No. of Species
	1	2	3	4		
Avinissery	66	211	109	16	402	56
Kanimangalam	66	37	139	115	357	35
Mannuthy	6	14	4	3	27	13

This study was conducted at different locations of Thrissur district. They collected and identified all together 84 species of hymenoptrans belonging to 60 genera and 21 families.

Important families of parasitic Hymenoptera

Narendran (2001) listed 48 parasitic families under Hymenoptera. Among which, 16 families (listed below) are found to be most common and abundant in rice ecosystem. They are:

Aphelinidae	Eupelmidae
Bethylidae	Eurytomidae
Braconidae	Ichneumonidae
Chalcididae	Mymaridae
Diapriidae	Platygasteridae
Elasmidae	Pteromalidae
Encyrtidae	Scelionidae
Eulophidae	Trichogrammatidae

Family Chalcididae contain the greatest species diversity. Families like Scelionidae and Trichogrammatidae are more common and consist of species having very high parasitic potential.

The major pests of rice are yellow stem borer, BPH, gall midge and leaf folder. The percentage yield reduction caused by them are given below:

Pest status of rice in India

Name of pest	Percentage yield reduction
Yellow stem borer	10-25
Brown plant hopper	100 (severe case)
Gall midge	50 (severe case)
Leaf folder	10-50

Hymenopteran parasitoids associated with these major pests are discussed below:

RICE STEM BORER AND HYMENOPTERAN PARASITOIDS

Rice stem borer is a key pest of rice, causing considerable yield reduction. The symptom of attack is manifested through the appearance of dead heart in early crop stage and white ear head after production of earhead.

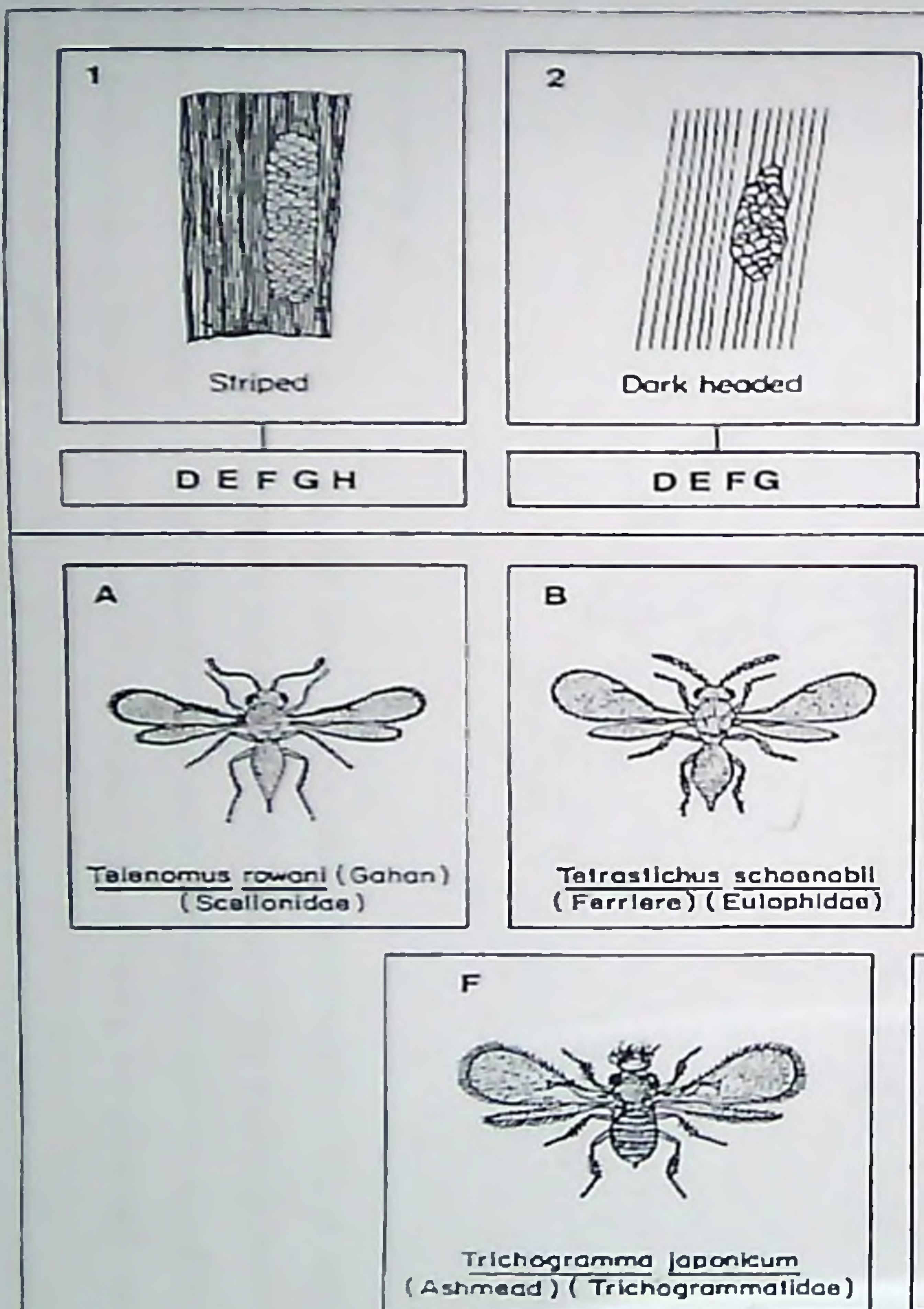
The egg mass of stem borer is covered with a thick tuft of hairs and each egg mass contains 15-80 eggs.

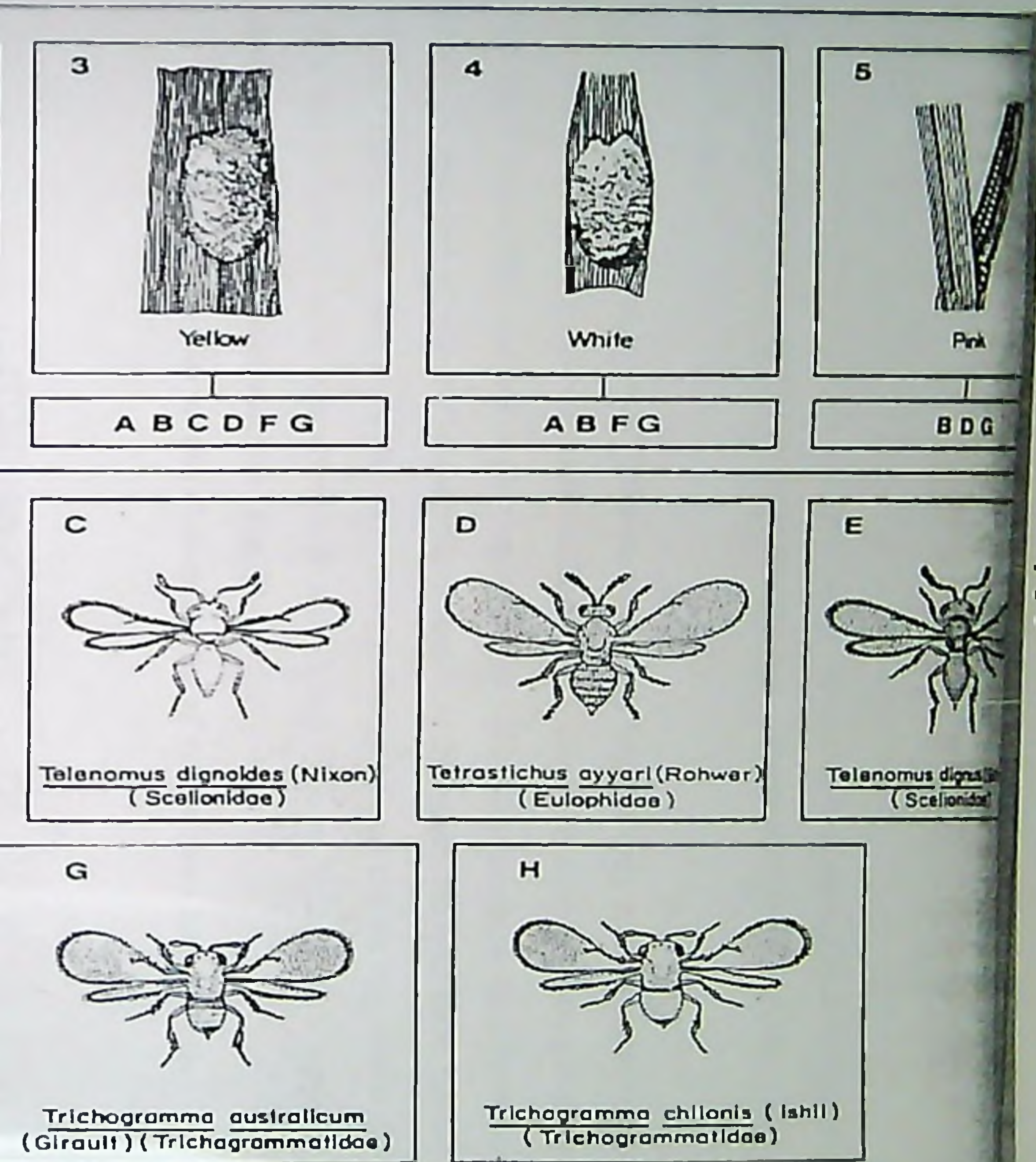
Egg parasitoids of rice stem borer

Eight egg parasitoids are found to be parasitising the different stem borer species either alone or in combination (Fig. 1). Most of the parasitoids are attacking the egg mass of yellow stem borer. Egg parasitism often goes beyond 90%.

Among the egg prasitoids the most commonly seen are the species of *Tetrastichus*, *Telenomus* and *Trichogramma*. Most often these parasitoids occur together in stem borer egg causing egg parasitism of 58% and egg mass parasitism of 75.3% (Reissig *et al.*, 1986).

Fig. 1. Egg parasitoids of rice stem borer




3
Yellow
A B C D F G
4
White
A B F G
5
Pink
B D G
C
Telenomus dignoides (Nixon)
(Scelionidae)
D
Tetrastichus ayyari (Rohwer)
(Eulophidae)
E
G
Trichogramma australicum
(Girault) (Trichogrammatidae)
H
Trichogramma chilonis (Ishii)
(Trichogrammatidae)

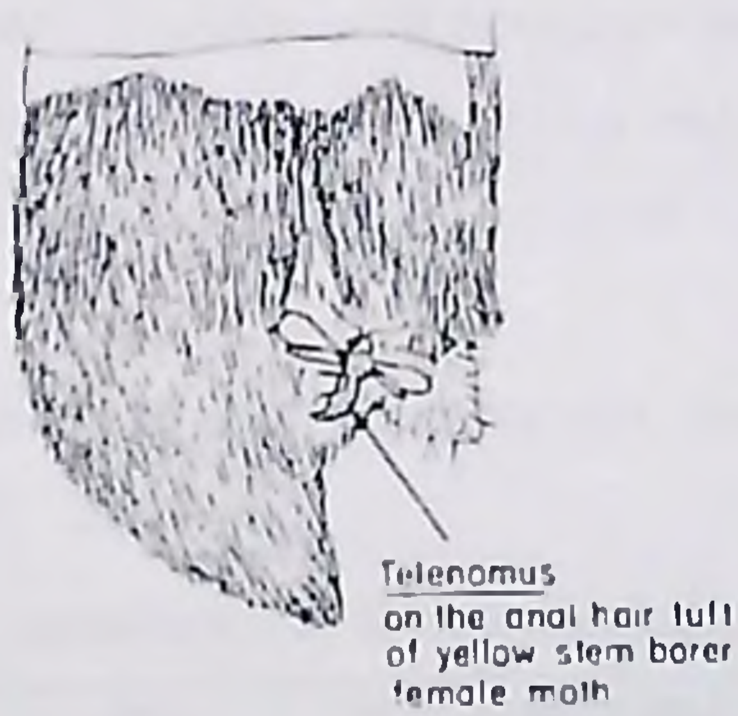
Tetrastichus sp.

Tetrastichus sp. is an eulophid wasp which has got strong and elongated ovipositor with which the wasp can penetrate the hairy covering of the egg mass.

Scelionid wasp

Scelionid wasp, i.e., *Telenomus* sp. which has got only a short ovipositor, shows another adaptation for oviposition. The wasp locates the female moth, possibly by the wind borne chemical or sex pheromone given off by the female moth. The wasp attaches the anal tuft of hair in the abdominal region of the moth and wait there till the moth lays eggs. Soon after the moth lays its egg, the wasp also put its egg above the moth's egg before the latter is covered with a thick tuft of hairs. That much adaptation is observed in parasitic Hymenoptera (Fig. 2).

Fig. 2. Scelionid wasp on the abdominal tip of yellow stem borer moth



Trichogramma sp.

Trichogramma sp. are the most widely and commercially utilised egg parasitoids of lepidopteran pests. Since this parasite kills he pest in the egg stage itself before the pest could cause any damage to the crop and also that it is quite amenable to mass production in the laboratories, it has the distinction of being the most utilised biological control agent in the world.

Mass production of *Trichogramma* spp.

In India, the eggs of rice moth, *Corcyra caphalonica* is generally used as a factitious host for the production of *Trichogramma* sp. The *Corcyra* moths are introduced into an oviposition cage fitted with wire mesh at the bottom. The eggs laid by the moths pass through the mesh and get collected in a box attached to the cage (Manjunath, 1988). These eggs are contaminated with moth scales, broken legs etc. Therefore these are passed through the sieve while the eggs and larger contaminants remain on the sieve. Then the eggs are to be sterilized under UV light. The sterilized eggs are taken in a vial and closed with a lid fitted with fine wire mesh.

Cards of required size are selected and one side of it is coated with a thin layer of diluted gum solution. Such gummed card is placed in a tray and the vial containing *Corcyra* egg is shaken all over it. Eggs fall through the mesh and get glued to the card in uniform layer. These egg cards are introduced into the cage containing parasitoids and thus the *Corcyra* eggs on cards are parasitised by *Trichogramma* spp.

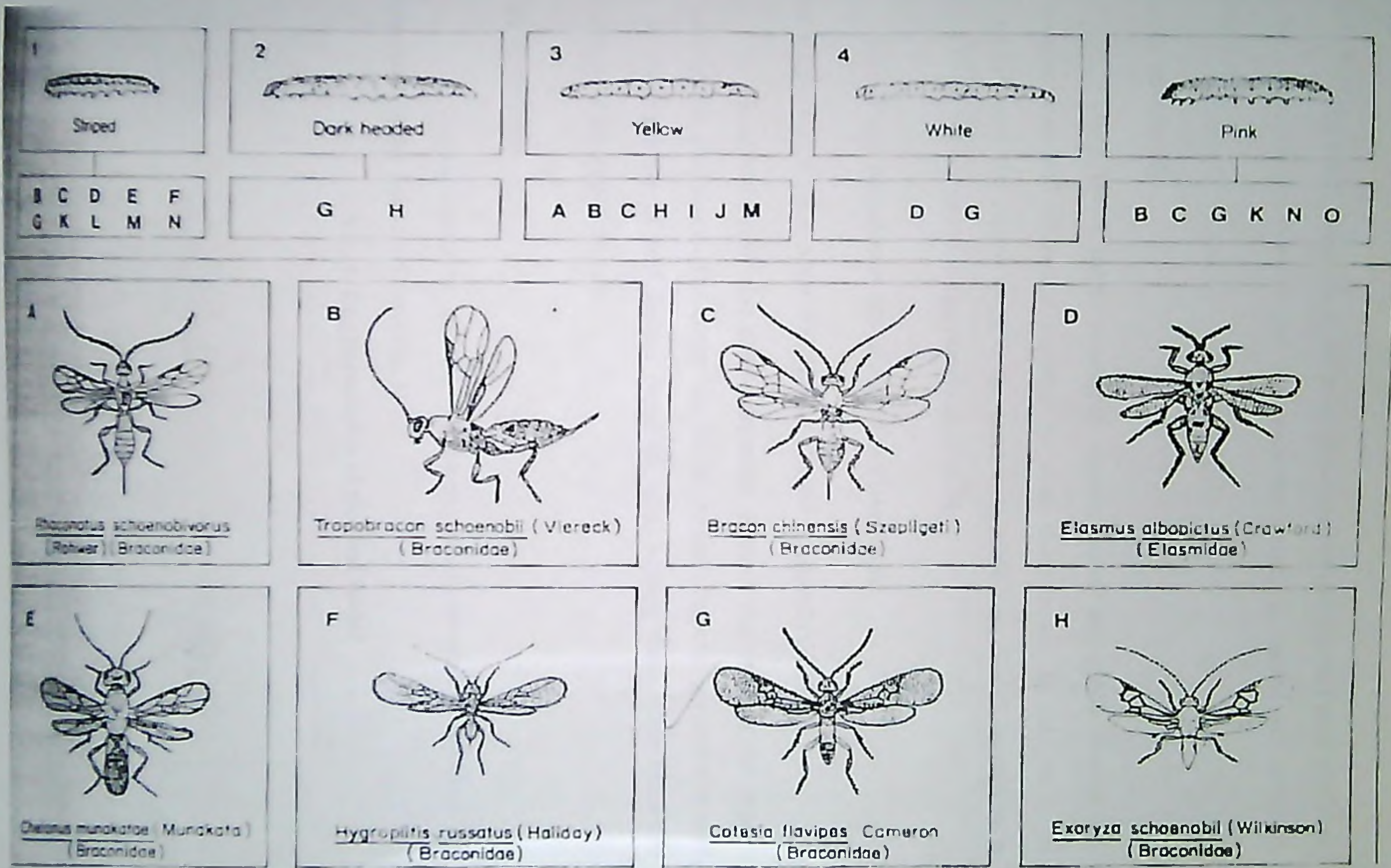
In rice ecosystem, *Trichogramma japonicum* and T. *chilonis* are mainly released against rice stem borer and leaf folder. An experiment was conducted at Assam Agricultural University to evaluate *T. japonicum* and *T. chilonis* against rice stem borer.

Two dosages of these egg parasitoids was tried along with need based spray of insecticides (PDBC, 2000).

Evaluation of *T. japonicum* and *T. chilonis* against rice stem borer

Treatment	Pre-released	% dead heart due to stem borer at weekly interval				% which ear heads	Yield kg/ha
		1	2	3	4		
T. japonicum + *T. chilonis* @ 50,000/ha	3.17	3.86	3.14	2.22	2.04	2.90	4373
T. japonicum + *T. chilonis* @ 1 lakh/ha	3.24	3.51	2.67	2.34	2.97	2.98	4107
Insecticide	3.30	4.22	3.91	2.53	2.39	2.18	4026
Control	3.51	4.99	4.71	8.00	8.35	7.56	3186

Fig. 3. Larval parasitoids of rice stem borer



The result showed that, the damage percentage was less and yield was high in the *Trichogramma* released plot as against the insecticide treated and untreated control plots.

Larval parasitoids

There are 8 larval parasitoids coming under the family Braconidae, Ichneumonidae and Elasmidae reported on stem borer larvae. In case of larval parasitoids also the yellow borer is the most susceptible host. Larval parasitism is found to be less compared to egg parasitism (Fig. 3).

Pupal parasitsm is rare in case of rice stem borer.

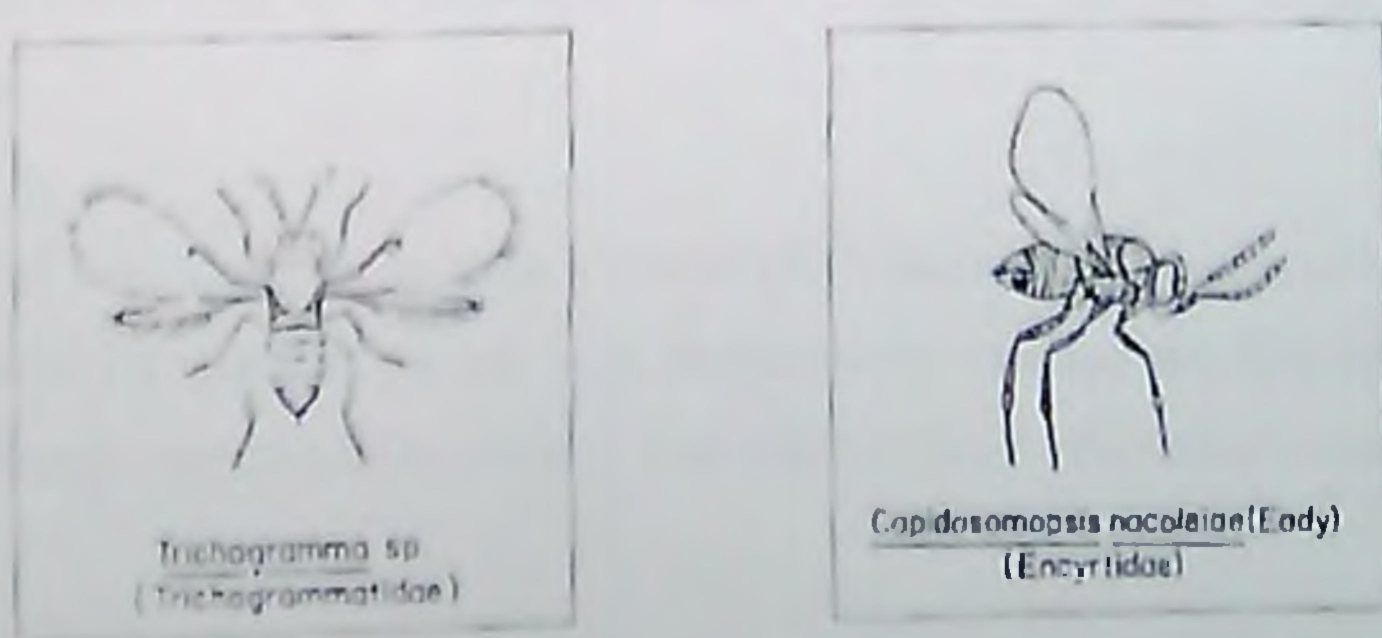
RICE LEAF FOLDER AND HYMENOPTERAN PARASITOIDS

Rice leaf folder also one of the major pests of rice infesting the crop in almost all the crop stages. The larvae fold the leaves and scrape the green matter with in it. Four species of leaf folders occur in Asia. Among these, *Cnaphalocrocis medinalis* is the important one.

Egg parasitoids

Trichogramma sp. and *Copidosomopsis nacoleiae* are the important egg parasitoids in the field condition (Fig. 4).

Fig. 4. Egg parasitoids of rice leaf folder



Trichogramma chilonis is mass multiplied and released in the field against leaf folder. Egg cards of blue colour is used against leaf folder whereas white cards are used against rice stem borer. Field evaluation of *T. japonicum* and *T. chilonis* was conducted against leaf folder also at Assam Agricultural University (PDBC, 2000).

Evaluation of *T. japonicum* and *T. chilonis* against rice leaf folder

Treatment	Pre-release population	Mean infestation of leaf folder at weekly intervals			
		1	2	3	4
T. japonicum + *T. chilonis* @ 50,000/ha	2.19	2.49	2.11	1.99	1.77
T. japonicum + *T. chilonis* @ 1 lakh/ha	1.68	2.78	2.54	2.05	2.28
Need based insecticide	1.33	2.86	3.06	2.19	2.06
Control	2.50	2.50	4.23	5.06	5.33

PDBC, 2000

Here also positive results were obtained i.e., the percent infestation of leaf folder was found to be low compared to the insecticide treated and untreated plots.

It is better to keep the two trichocards (white card of *T. japonicum* and blue card of *T. chilonis*) together in the field. Otherwise, we have to take chemical control measures against rice leaf folders, which will destroy the population of *T. japonicum*. If we keep the two cards together, the population of the two pests is maintained at lower levels and we can apply need based pesticides in the initial stages.

Works done in Kerala using *T. japonicum* and *T. chilonis* against rice stem borer and leaf folder

1. The results of four years study from 1993 to 1997, during rabi season revealed that *Trichogramma* release was on par with untreated control for the first three years and in the fourth year chemical control was inferior to all other treatments (Lyla *et al.*, 2000).

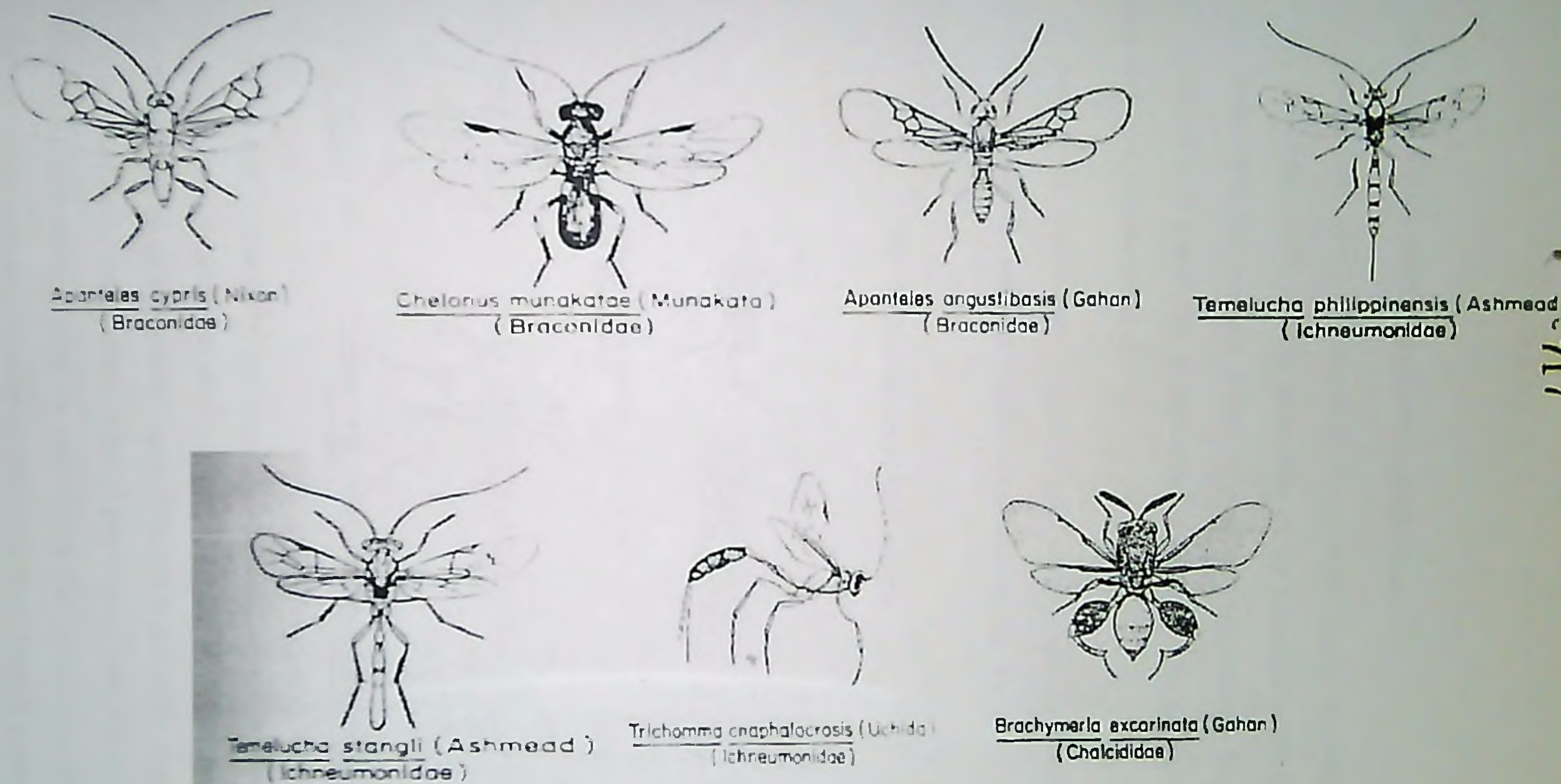
2. Another experiment was conducted with higher doses of *T. japonicum* and *T. chilonis* @ 50,000/ha and 1 lakh/ha for 3 years. Observations were recorded on the infestation by stem borer and leaf folder and the percentage dead hearts, white ear heads and leaf folder damage. Yield parameters like total weight and grain weight were also recorded. Pooled analysis of the data for three years indicated that no significant difference was there amongst the *Trichogramma* released and insecticide treated plots. This results indicate the effective role played by natural biocontrol agents in the paddy ecosystem of Kerala (KAU, 2000).

3. A field experiment was conducted during rabi, 2001 in Palakkad district in Kerala to evaluate the effectiveness of methods for rice pest management with particular reference to stem borer and leaf folder. The inundative release of egg parasitoids *T. chilonis* and *T. japonicum* at two dosage levels @ 50,000 and 1 lakh/ha and *Bacillus thuringiensis* were tested along with need based chemical control treatment and untreated control. Six releases of *T. japonicum* and *T. chilonis* @ 1 lakh and 50,000/ha at weekly interval starting from 15 DAT could reduce the white ear head infestation to the extent of 69.88% and 55.59% respectively as compared to control. Grain yield was high in *Trichogramma* released plots @ 1 lakh/ha the percent increase being 20.3% as compared to untreated control and 11.8% as compared to chemical control. It was found that six releases of *T. chilonis* and *T. japonicum* @ 1 lakh/ha is better than chemical control in managing stem borer infestation and increase in yield in rice (Beevi *et al.*, 2002.)

Larval and pupal parasitoids of leaf folder

Species belonging to Braconidae, Ichneumonidae and Elasmidae are the larval parasitoids. Larval parasitism is found to be 37%. The important pupal parasitoid is *Brachymeria excarinata* (Chalcididae). Pupal parasitism in the field is 15-44% (Fig. 5).

Fig. 5. Larval and pupal parasitoids of leaf folder

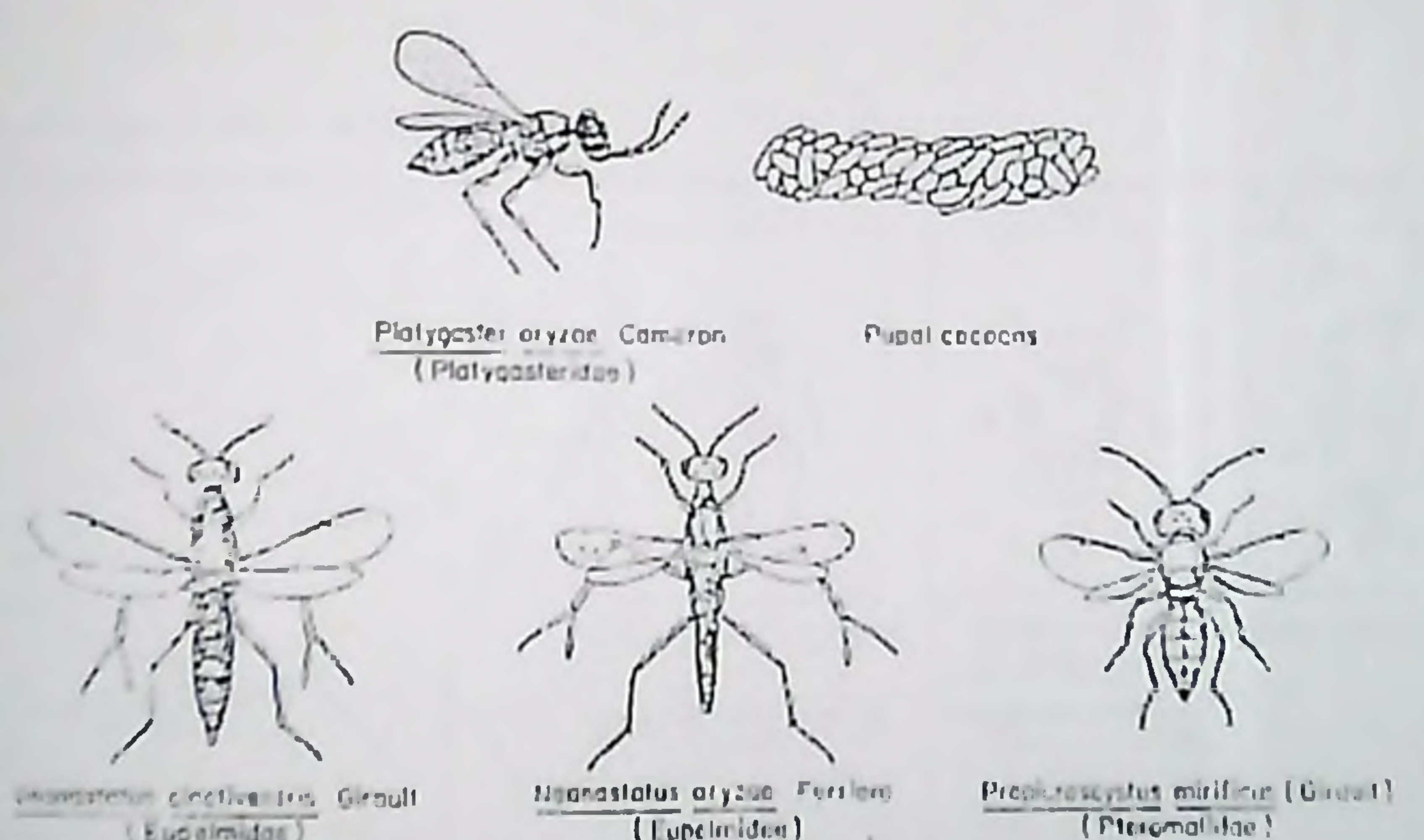


GALL MIDGE AND HYMENOPTERAN PARASITOIDS

Gall midge is also a serious pest of rice especially during the tillering phase of the crop. The midge causes damage to the crop by the formation of the gall in the place of the central leaf. The presence of the pest is noticed only after the formation of the gall. Application of insecticide is ineffective at that time. So biocontrol is the alternative and more suitable method.

Among 19 parasitoids attacking gall midge, species belonging to Platygasteridae, Eupelmidae and Pteromalidae are important (Fig. 6).

Fig. 6. Parasitoids of Gall midge



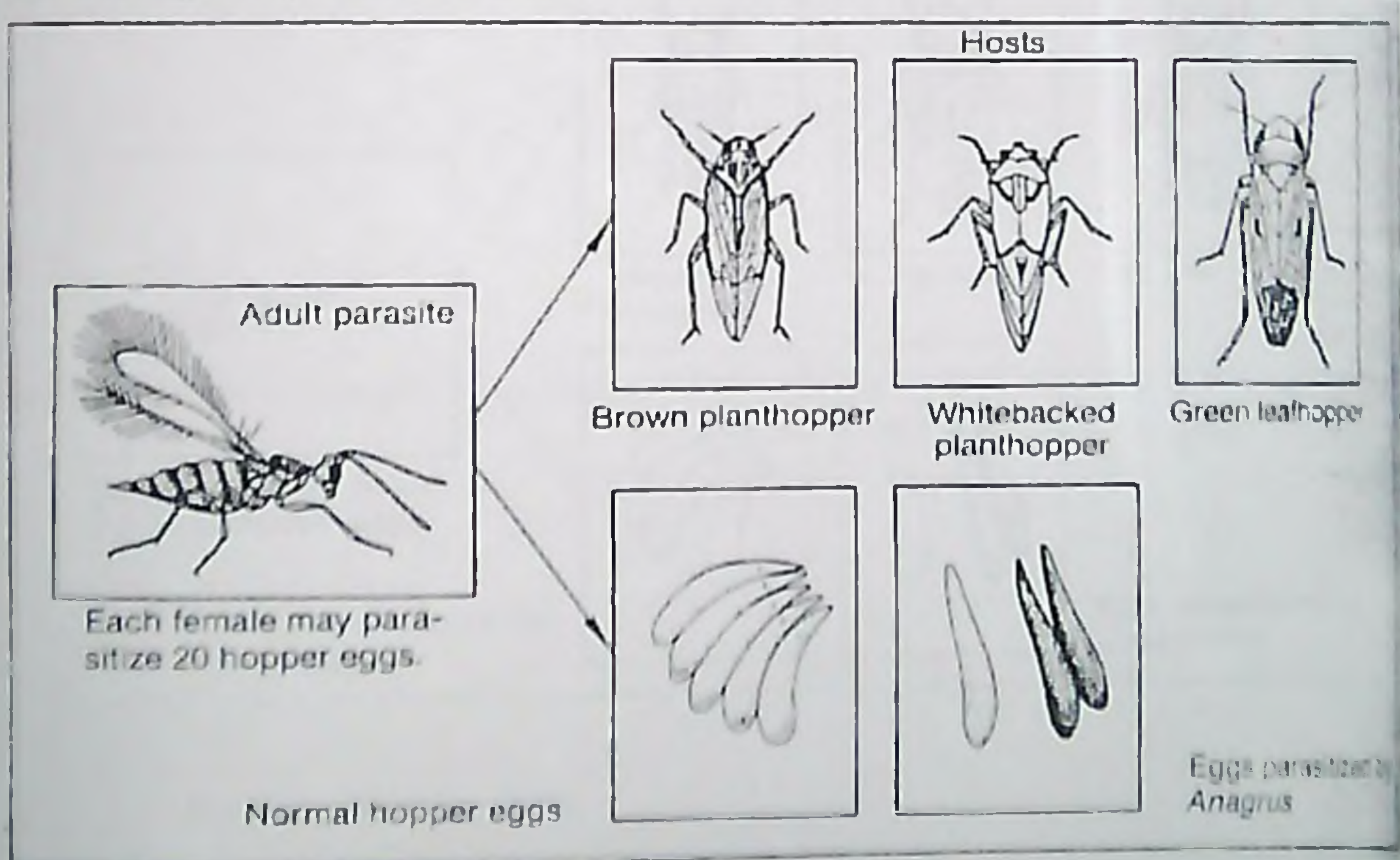
Platygaster oryzae is the key parasitoid attacking the egg and larval stages of the pest. The parasitism have been observed to be low early in the season, 40 to 50 per cent in the tillering phase and reaches up to 95% at the end of the season. Field studies have revealed a consistent pattern of fall in pest damage with concomitant rise in parasitisation. Hence there is need for augmenting the parasitoid population to manage the pest in the early stage through inundative release. Though a technique for mass multiplication of gall midge is available, efforts to produce the parasitoid, *P. oryzae* for field release are lacking.

PLANT HOPPERS, LEAF HOPPERS AND THEIR PARASITOIDS

Brown plant hopper and Green leaf hoppers are the serious pest of paddy cultivation. They cause direct damage by sucking the sap. More over, indirect damage is caused by acting as vectors of virus diseases of rice.

The eggs of hoppers are parasitised by about 17 parasitoids belonging to the families Mymaridae, Trichogrammatidae and Scelionidae. Egg parasitism considerably varies among hopper species. Mymarid only constitute about 82.4% to 97.6% of total parasitoid attacking the hoppers. *Anagrus* sp. and *Gonatocerus* sp. are the important among them (Fig. 7 and 8).

Fig. 7. *Anagrus* sp. and their hosts



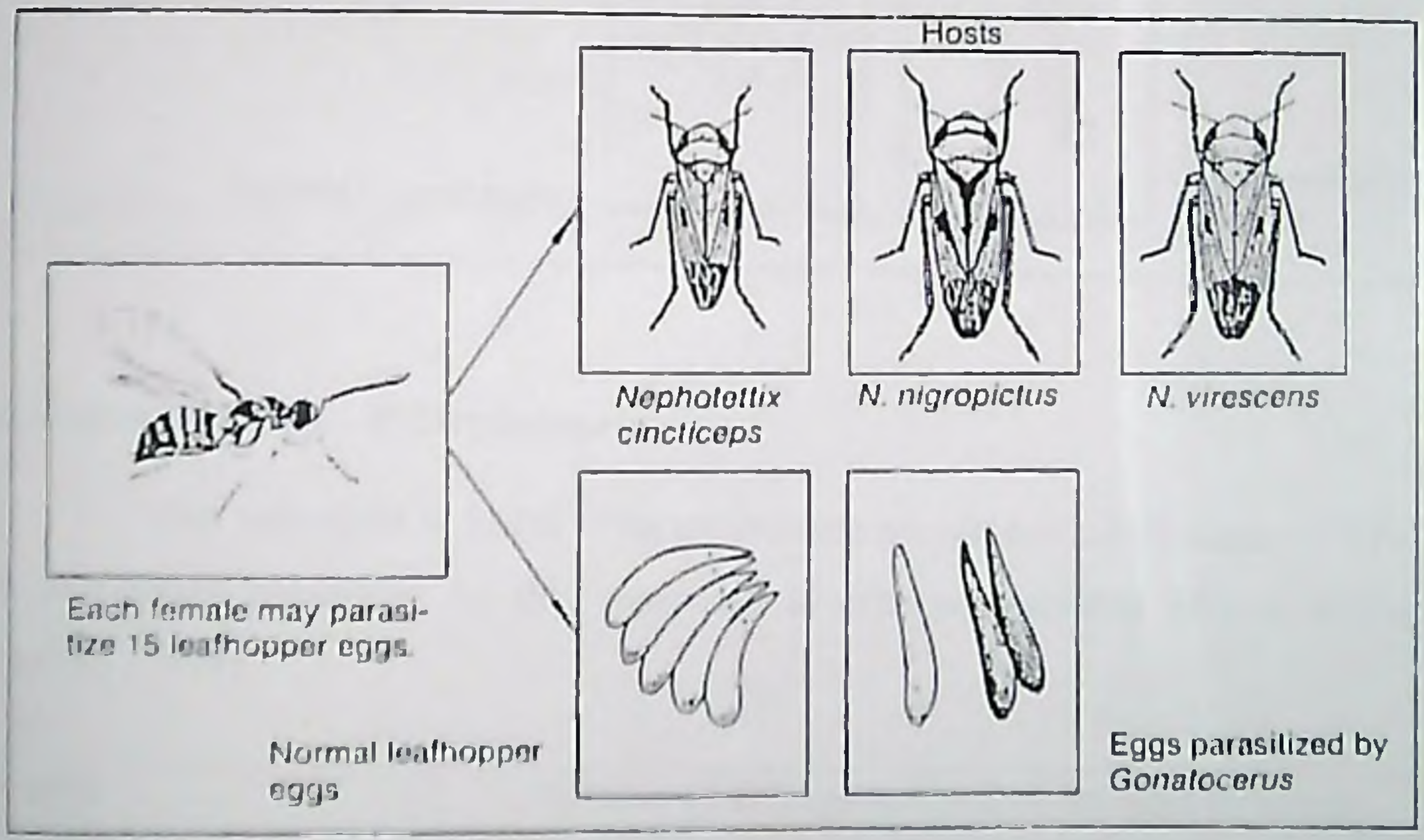
Percent parasitism by *Anagrus* sp. is about 64%. Under green house condition, biocontrol potential of *Anagrus* sp. was studied against BPH (Beevi *et al.*, 2001).

Influence of mymarid parasitoid (*Anagrus* sp.) on hatching of *Nilaparvatha lugens*)

Treatment	% of reductions in viability
Control	-
1 pair of parasitoid released from day 5 to 15	59.5
5 pairs of parasitoids released from day 5 to 15	84.9

From the table, it is clear that there is a positive co-relation exist between the parasitoid population and the percent reduction in egg viability.

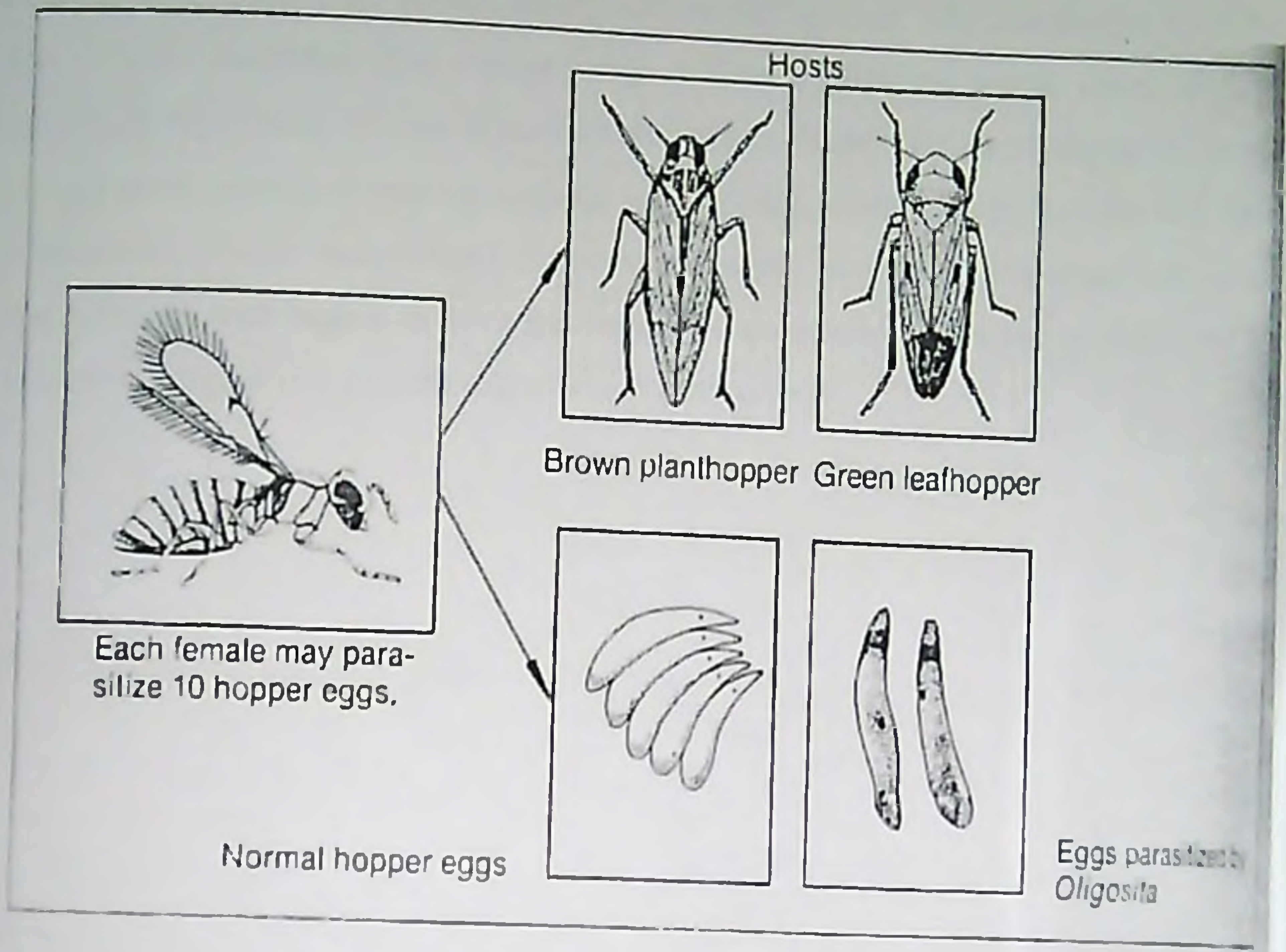
Fig. 8. *Gonatocerus* sp.



Percent parasitism 40%.

Oligosita sp. is a Trichogrammatid parasitoid. Generally Trichogrammatids parasitise the eggs of moth, this is an exceptional case. Percent parasitism is 55% (Fig. 9).

Fig. 9. Oligosita sp.



Pseudogonatopus sp. - F. Dryiinidae

This parasitoid is found to be parasitising nymph and adult stages of BPH. The percentage parasitism by this parasitoid in different varieties of rice studied (Bhaskar, (1999).

Varieties	Number of BPH parasitised at two host densities	
	10	20
Jaya	2.12	3.64
Jyothi	1.92	3.96
Kanakom	1.88	3.00

Variety Jyothi shows higher percentage parasitism compared to others.

CONCLUSION

Hymenopteran parasitoids show a very high species richness and abundance in most of the ecosystem.It is observed that from Thrissur district itself 84 species were identified. You cannot imagine, from a single net sweep itself, we will get a high population. In case of parasitoids such as *Platygaster* sp., *Anagrus* sp., even though some laboratory rearing methods are available, their mass production and field release are yet to be standardized. Conservation aspect is the most important biological control tactic with regard to these hymenopteran parasitoids. This can be achieved by selective, rational and discriminative use of pesticides.

DISCUSSION

1. What is the price of trichocard? From where it is available?

Trichocards are available at a price of Rs.15/card from Biological Control Lab., Mannuthy. It is also available from Project Directorate of Biological Control (PDBC), Bangalore.

2. Give the dosage and frequency of *Trichograma* release in rice?

The recommended dose is 1 lakh egg or adult per ha. In each card there are 18,000 to 20,000 eggs. So we require 4 or 5 cards per hectare. The release should start from 30 to 45 DAT and five such release is to be done at weekly interval for effectiveness. The trichocards, after splitting in to small pieces are stapled on the leaves. During rainy season it is better to keep the cards in a plastic container, one end of which is covered with a wire net. The plastic container is to be tied on a stick placed in the field.

3. There is no significant difference among the *Trichograma* released and chemical treated plots in Kerala condition. What is the reason?

In Kerala, rice ecosystem is enriched with a great diversity of Hymenoptran parasitoids and other natural enemies. So there is a very much scope for effective natural biocontrol. Hence the applied treatment has no effect in the yield of crop.

4. What is *Eriosoma lanigerum?*

It is woolly aphid of apple.

5. Can we take biocontrol method alone for the control of major pests of rice?

Due to the indiscriminate use of pesticides, the rice ecosystem is the most spoiled ecosystem. So we cannot get the benefit of biological control with in a shorter period. Gradually reduce the pesticide application, try to enhance the natural enemy population, there by we can surely shift on biocontrol alone. More over this should be taken as a group approach.

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PHYSIOLOGICAL RESPONSES OF BOTANICALS IN INSECT SYSTEMS

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SEMINAR REPORT

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ABSTRACT

Recently prime importance is given to IPM practices in crop protection. It emphasizes the need for simpler and ecologically safer methods to reduce the environmental pollution and other problems like toxicity to non-target organisms, development of resistance and the search in this aspect leads to the identification of several plants, which can be used as safe and renewable source of insecticide.

Botanicals offer good promises in pest- management, as they are soft, naturally occurring and with no or very little adverse environmental impacts. Many botanicals as pesticides are available all over the world for the control of different harmful pests. Popular botanicals are pyrethrum and pyrethroides (from *Chrysanthemum cinerariaefolium*), rotenone and rotenoids (from *Derris elliptica*), nicotine and nicotenoids (from *Nicotiana tabaccum* and *N. rustica*) and various neem principles like azadirachtin (from *Azadirachcta indica and Melia azadirach*). Responses of these botanicals on insect physiology are of different types, which should be properly known for their practical exploitation in IPM.

Antifeedants are chemicals that inhibit feeding in insects. Antifeedant activity in neem, against *Nephotetix virescens*, was detected by Saxena and Khan (1985). Botanicals can act as protein or cuticulin inhibitors on insects. Azadirachtin and plumbagin on exposure showed disturbance in cuticulogenesis visible externally as unsclerotized incompletely tanned areas in pupal state (Krishnayya and Rao, 1995) of some insects

An evaluation of the insect growth inhibitory activity of essential oils of some medicinal plants was taken up by Sharma *et al.* (2001). The ovipositional deterrency of extracts of *L. aspera, O. sanctum, etc.* against the mosquito was assayed by Murugan and Jeyabalan (1999). Major problems of this botanicals are the lack of confidence in such products due to inconsistency of results, lack of standardization for biological efficacy and their photodegradable nature. Further development in these low cost technologies will be very useful for the poor farmers in practicing the acclaimed IPM technology.

INTRODUCTION

Recently prime importance is given to the phenomenon of IPM in all fields of agriculture. The system utilizes all suitable techniques and methods in as compatible a manner as possible to bring the pest population at levels below those causing economic injury. IPM emphasizes the need for simpler and ecologically safer methods for pest control so as to reduce the environmental pollution and other problems caused by excessive and indiscriminate use of pesticides as, toxicity to non target organisms, development of resistance, environmental contaminations and their ever increasing cost. The search for environmentally sound methods to manage insect pests is therefore, being carried out all over the world. The plant kingdom affords a rich storehouse of chemicals of diverse biological affects on insect. In recent years several plants have been identified which can be used as safe and renewable source of insecticide.

Plant based insecticides or botanical pesticides or botanicals offer good promise in pest- management, as they are soft, naturally occurring with no or very little adverse environmental impact. Many botanicals as pesticides are available all over the world for the control of different harmful pests. Further development of low cost technologies will be very useful for the resource poor farmers.

Botanicals are pesticides made from active compounds found in certain plants. They are plant secondary compounds, usually alkaloids that have insecticidal properties. Plant yields the following products that are used in insect control. They are repellents, toxicants, attractants, solvents for insecticides and dust carriers. Recent research in US and Canada to find other naturally occurring active compounds in plants has come up with many promising plant products.

A major problem associated with large - scale adoption of botanical pesticides is the lack of confidence in such products due to inconsistency of results. The derivatives would require some degree of standardization for biological efficacy. As the extracts are photodegradable, better timing of application is also important. Another problem relates to farmer's perception. But the farmers of our country are well aware of the beneficial effects of botanicals. A number of commercial formulations are now registered in India. Among the popular botanicals are pyrethrum, rotenone, nicotine and azadirachtin

Advantages of botanicals

- Rapid breakdown of materials, there will be little or no risk of residue on food crops
- Some materials can be used before harvest
- Acts quickly to stop feeding and they may not cause death for days
- Few exceptions of botanicals have low to moderate toxicity.
- Action as stomach poisons and rapid degradation can be more selective against pests rather than beneficial species.
- Are non-toxic to plants.

Disadvantages

- Creates need for precise timing or more frequent application.
- Tend to cost more than synthetic insecticides and are non readily available.
- Potency often varies from among batches

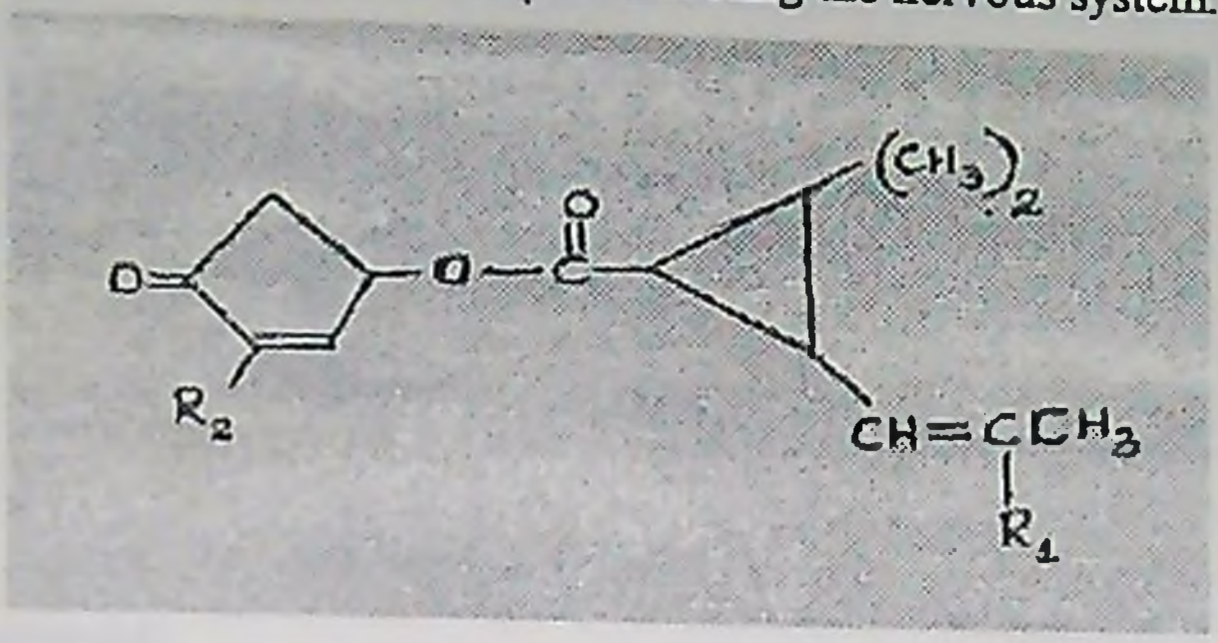
COMMERCIALLY USED BOTANICALS

Pyrethrins & Pyrethroids

Pyrethrins are one of the most extensively used botanicals in the past. The active ingradients in Pyrethrum are extracts from the Chrysanthemum *Chrysanthemum cinerarifolium.* Most flying insects are highly susceptible to pyrethrins, causing them to 'drop' almost immediately upon exposure. This is comparatively safe to mammals, being harmful against only to those who are allergic to it. Its allergic property is stated to be due to the presence of certain impurities and the pure extracts are devoid of this effect.

The active ingredients of pyrethrins are six esters. Pyrethrum I and II, Cinerin I and II and Jasmolin I and II whose structural formulae are similar. LD_{50} – 200

Pyrethrin is a quick knock down insecticide and acts on insects central nervous system causing immediate paralysis. Another popularity is its fast knock down household aerosols. However unless it is formulated with one of the synergists, most of the paralysed insects recover and ones again become a pest. It is unstable in sunlight. It is a contact insecticide affecting the nervous system.



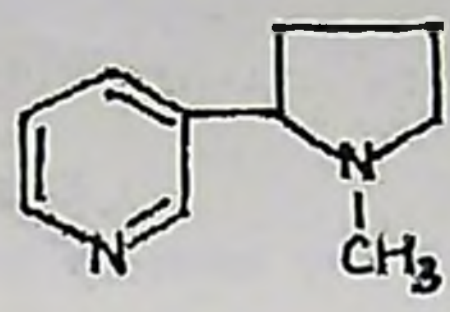
Structure of Pyrethrin

Pyrethrum has been formulated as dust by grinding the dried flower and mixing with a powder or with a known alkaline carrier as spray by extracting the active materials in solvent and aerosols.

Nicotines & Nicotinoids

It is an organic compound, which is the principal alkaloid of tobacco. Nicotine occurs throughout the tobacco plant with its concentration being more in leaves. The compound constitutes 5% of the plant by weight. Both the tobacco plant *Nicotiana tabaccum* and *Nicotiana rustica* are used for extraction. Nicotine is one of the liquid alkaloids. The alkaloid analogues are Nornicotine, Anabasin, Nicotine, and Cotinine etc. Among these, Nicotine is commonly used as insecticides. LD_{50} – 50.

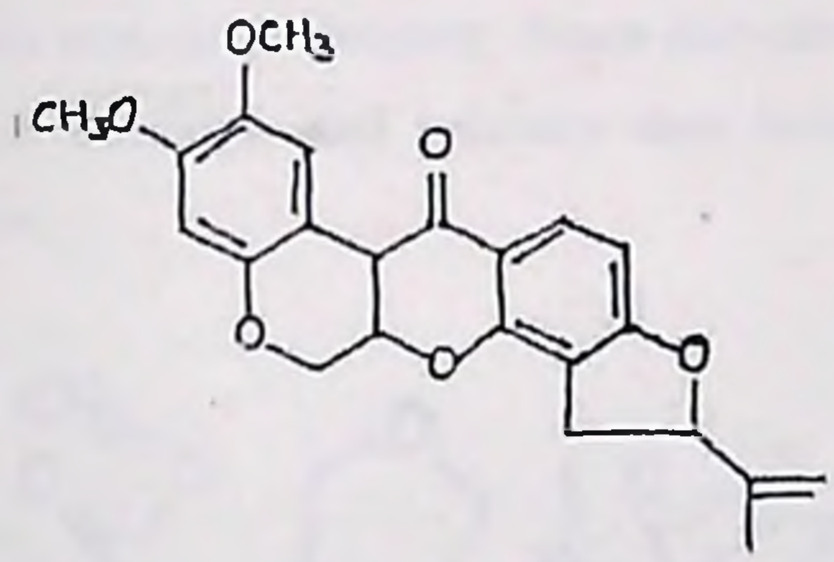
It has low toxicity to plants than to insects. In insects, it acts as a contact and stomach poison affecting the nervous system. Nicotine is active by virtue of binding acetylcholine receptor protein on the postsynaptic membrane of the cholinergic synapse. Nicotine mimics acetylcholine at the neuro-muscular junction in insects. It acts as an agonist thereby binding, activating or stimulating the receptor. The action produces hyperactivity twitching, convolutions and death, all in a rapid order. In insects some action is observed only in the central nervous system ganglia. A 40 % solution of nicotine was ones used as garden insecticides. Targeted organisms are soft bodied sucking insects like whitefly, thrips and bollworm in cotton, brown planthopper and green leafhopper in rice; grubs in brinjal, potato and cauliflower.



Structure of Nicotin

Rotenone & Rotenoids

It is an alkaloid extracted from the roots of many tropical legumes such as *Derris elliptica, Lonchocarpus nicou* and *Tephrosia vogelii.* The plants have been used since ancient times to kill or paralyse the fish. Roots of these plants are used to extract rotenone. LD_{50} –132.



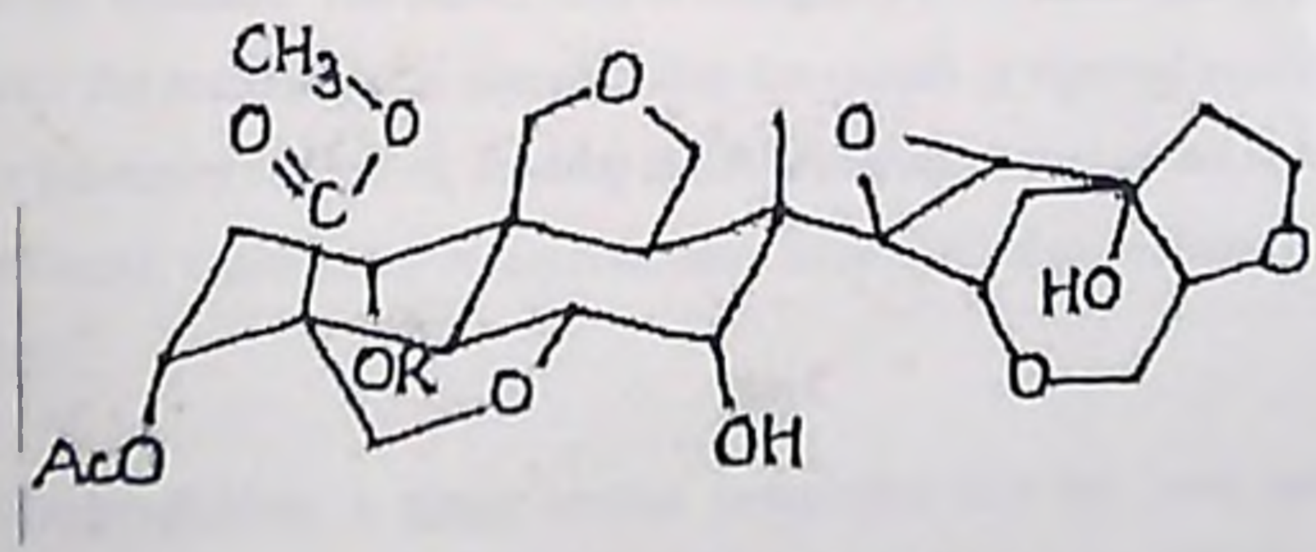
Structure of Rotenone

It is both a stomach and contact insecticide used for the control of leaf eating caterpillars. Rotenone has recently been linked to Parkinson's disease. It affects respiration by inhibition of oxygen utilization by body cells resulting in the death of the insects.

Mode of action of rotenone is at the phosphorylation step of respiration in mitochondria. Because of this, rotenone is a general poison to all animals, which explains its high toxicity. Typical symptom of rotenone poisoning in insects are depressed heartbeat and respiration. It also acts by inhibiting one of the oxidation steps in the mitochondrial electron transport system of ATP production. Rotenone rapidly degrades in sunlight and so has limited use in crop protection.

Neem Derivatives

Derived from seeds of the neem tree, *Azadirachcta indica.* Neem has assumed the status of an international tree. The overall knowledge concerning the efficacy and the biological property of neem is far from complete. Neem is bitter in taste due to the presence of an array of complex compound called triterpenoids specifically limnoids. At present, 10 limnoids that have been identified are Salannin, Salannol, Salannol acetate, 3-Diacetyl salannin, 14-Epoxy azaradion, Gedunin, Nimbinen, Diacetyl nimbinen, Azadiradion and Azadirachtin. Of these, azadirachtin is the most active compound. It is a steroid like tetra terpenoid comprising seven isomers AZA to AZG. The AZA is the most important compound in terms of quantity (83%) and insecticidal property. Neem derivatives do not kill the insects. They modify their behaviour and influence their biological process in a very detrimental manner.



Structure of Azadirachtin

Other plants used for the extraction of the botanicals

Yellow oleander, Acorus, Ageratum, Ryanodine, Quassin, Sabadilla, Pellitorine

PHYSIOLOGICAL EFFECT OF BOTANICALS IN INSECT SYSTEMS

Antifeedant or phagodeterrent effect

Antifeedants are chemicals that inhibit feeding in insects when applied on the foliage without impairing their appetite and gustatory receptors or driving them away from the food. They are also called phagodeterrents or gustatory repellents or feeding deterrents or rejectants. With these insect dies slowly.

Advantages:

- Affects only the polyphagous insects and so do not act upon the beneficial ones.
- Produce no phytotoxicity and pollution

Disadvantages

- Only chewing type of insects are affected by this
- Insects are not killed immediately, they move to untreated parts or other plants and damage them too

In order to have a sustained feeding an insect requires:

1. Presence of gustatory stimulus
2. Absence of inhibitory stimulus
3. Presence of hunger

Antifeedants interfere with the taste receptors of mouth region, lacking the right gustatory stimulus. The insect fails to recognize the treated leaf as food. This is proved that if the antifeedant is placed within the mouth or injected into the blood by passing the gustatory receptors, feeding is not prevented because the receptors have not been affected. Quite many researches have been carried to evaluate botanicals as antifeedants.

Andrographolide, a major known compound that has been isolated from *Andrographis paniculata* an indegenous herb in South East Asia, effectively reduced larval feeding (Moktadar & Sincar 1939; Chakravarthi and Chakravarthi, 1952)

As early as 1975, Sandhu and Singh reported the extracts of neem seed to inhibit feeding in some field pests like *Pieris brassicae and Chrotogonus* *trachypterus*. Antifeedant insecticidal activities following the application of neem compounds has also been studied by Rembold *et al.* (1982).

As for plant chemicals the antifeedant activity in neem, *Azadirachta indica* against *Nephotetix virescens*, the most effective vector of tungro disease was detected by Saxena and Khan (1985). Chouhan and Qadri (1989) observed 88% protection of okra fruit against fruit borer with 1000 ppm of datura extract.Again, neem isolates have been reported to possess antifeedant properties against *Spodoptera litura* (Ayyangar and Rao,1989).Azadirachtin injected into the 6^{th} instar larvae at 1 / g significantly reduced the consumption , digestion and utilisation of dry matter and carbohydrate, but the conversion of nitrogen to dry matter was in treated than in untreated larvae. Azadiractin resulted in lower utilisation of nutrients.

Assesment of the effects of methanol extracts of *Ocimum sanctum Linn*. on the semilooper, *Anomis sabulifera* Guen.for its antifeeding property (Mallick and Banerji, 1989). Antifeedin propertynof each conc. Was judged comparing the area of leaf consumed by one pair of larvae with that of control by measuring the area with the help of planimeter after allowing to feed upto 72 hr. Consumption of leaf area was also recorded at every 24 hr .It was observed that the area of leaf consumed with 10 and 5% conc. Was nil up to 72 hr. In lower conc. Of 2% consumption of leaf area was almost nil upto 48hr. Antifeedant effect was also very promising upto 24 hr in the leaves treated with 1% conc. Insecticidal effect of the extract was nil with all conc.

In a study on the antifeedant activity of some plant products against the leaf caterpillar *Selepa docilis* Butl. on brinjal and wooly bear *Pericallia ricini* F. on castor ,Jacob and Sheela (1994) reported only the neem extract to exhibit antifeedant property against both *P. ricini* and *S. docilis*..(Table 1&2)

Table 1. Effect of plant extracts on the feeding of *Pericallia ricini* F. on caster

Name of the plant	Concen -tration %	Percent leaf area consumed					Feeding ratio	Rating
		R1	R2	R3	R4	R5		
Datura	3	27.67 (31.76)	43.93 (41.50)	35.73 (36.69)	38.34 (38.23)	50.26 (45.17)	41.78	+ + +
Datura	5	48.15 (43.97)	36.46 (37.17)	46.01 (42.71)	46.82 (43.14)	31.45 (34.14)	37.98	+ + +
Chromo-leana	3	48.23 (43.97)	24.84 (29.87)	56.78 (48.91)	32.50 (36.76)	44.00 (41.55)	46.64	+ + +
Chromo-leana	5	39.91 (39.17)	48.50 (49.89)	50.84 (45.46)	62.14 (52.00)	21.84 (27.83)	41.26	+ + +
Neem	3	28.48 (32.27)	28.86 (32.52)	28.13 (32.03)	13.35 (21.47)	20.50 (26.92)	23.89	+ + +
Neem	5	16.74 (24.12)	26.31 (30.85)	21.32 (27.49)	12.96 (20.05)	13.61 (21.64)	18.29	+ + + +
Calotro-pis	5	25.68 (30.45)	34.26 (35.79)	42.16 (40.46)	19.82 (26.42)	22.66 (28.45)	28.91	+ + +
Control		100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)		

CD 5% = 7.39, Figures in paranthesis are angular transformed value

Table 2. Effect of plant extracts on the feeding of *Selepa docilis* Butl. on brinjal

Name of the plant	Concen -tration %	Percent leaf area consumed					Feeding ratio	Rating
		R1	R2	R3	R4	R5		
Datura	3	40.70 (39.64)	28.30 (32.14)	61.15 (51.41)	35.44 (36.51)	32.43 (34.70)	39.61	+ + +
Datura	5	12.10 (20.36)	12.97 (21.13)	12.27 (20.53)	18.18 (25.25)	33.29 (35.24)	17.78	+ + + +
Neem	3	6.08 (14.30)	11.93 (20.18)	6.49 (14.77)	11.58 (19.91)	6.25 (14.50)	8.46	+ + + +
Neem	5	11.18 (19.55)	7.76 (16.22)	8.60 (17.05)	0.069 (4.76)	1.27 (6.55)	5.91	+ + + +
Control		100 (90)	100 (90)	100 (90)	100 (90)	100 (90)		

CD 5% = 30.07
Figures in paranthesis are angular transformed values

In a screening trial of natural products for insect antifeedant activity, Tripathi and Singh (1994) observed that methanolic extract of 37 plants botanically identified belonging to 21 families had feeding deterrancy against *Spilosoma obliqua* Walker. The extracts from *A. paniculata* has been found to have antifeedant properties against the first and fourth stadium larvae of Diamond back moth (Hermawan *et al.*, 1993, 1994)

Hazra (1994) also reported the antifeedant property of Lantana and *Pongamia pinnata* against aphids and other insect pest.

Nair (1996) determined the feeding deterrent effect of *A.calamus* extracts on melon fly *Bactrocera cucurbitae* (Coq.) by recording the no. of flies alighting to feed the food material sugar crystals, treated with the extracts. No choice and multiple choice tests were conducted each using aqueous and solvent extract. The number of floes alighting for sustained feeding on treated an untreated material was recorded for 10 minutes. The alightment for sustained feeding was forced to be considerably affected by treating the food with extracts, thus showing the feeding deterrent activity of the extract (Table 3).

Table 3. Mean number of melon flies (*B.cucurbitae*) alighting to feed on sugar crystals treated with graded doses of *A.calamus* extracts

Treatments	Dose (%)	Mean number of melon flies alighting to feed* (in 10 minutes)	
		No choice test	Multiple Choice test
Aqueous extract FW1	0.10	8.08b	1.84b
Aqueous extract FW2	0.50	6.92c	0.60c
Aqueous extract FW3	1.00	6.32c	0.20cd
Aqueous extract FW4	5.00	4.12d	0.20cd
Aqueous extract FW5	10.00	2.04e	0.16d
Aqueous extract FW6	Control	8.96a	6.08a
CD		0.73	0.41
Methanol extract FS1	0.10	3.40b	1.28b
Methanol extract FS2	0.25	2.28c	0.44c
Methanol extract FS3	0.50	1.68d	0.16c
Methanol extract FS4	1.00	0.64e	0.08c
Methanol extract FS5	5.00	0.16e	0.08c
Methanol extract FS6	Control	7.92a	7.12a
CD		0.55	0.52

Treatment means followed by common letters do not differ significantly at 5% level

*1g of sugar treated 1ml of extract

Antifeedant effect of the extract was evidenced by the reduced alightment of flies on treated food material were there is a definite reduction in mean alightment values of flight with increase in concentration of the extract both in the case of aqueous as well as solvent extracts

Study was undertaken to evaluate the effects of neem leaf exudate from reddish terminal leaves of neem *A. indica* on antifeedant property to *S. litura* (Kumar *et al*, 1997). The response of insect to neem allelochemicals was evident with tremendous antifeedancy in a dose depended manner. At 10% treatment, pronounced antifeedancy was observed. This observation is in agreement with Ganeshan *et al.* (1995) were many neem formulations were effective against *Earias vitella, Helicoverpa armigera* and *S. litura* were lesser feeding due to neem formulation application was evident.

Effect of certain plant extracts on the consumption of food by *S. litura* was evaluated (Chithra and Rao, 1996). The order of efficacy of plant extracts with respect to reducing the food consumption was NSK (5%) and *N. odorum* (5%) and *A. mexicana* (1%).

Studies were undertaken to determine the antifeedant effect of *Melia azadirach* and *Adathoda vasica* in tea mosqito bug by Deka *et al.* (1999). The results revealed that both the plants with chloroform, methanol and petrolium ether extracts showed antifeedant property. The first instar nymph produced 179.5of spots after 24 h and the nymphs treated with *M. azadirach* chloroform extract at 5% the same produced only 20 spots (Table 4).

Table 4. Effects of plant solvent extracts on feeding of tea mosquito bug

reatment	% Concen-tration	Chloroform No. of spots after			Petroleum ether No. of spots after			Methanol No. of spots after		
		24h	48h	72h	24h	48h	72h	24h	48h	72h
Melia zadirach	5.0	20.0	30.0	45.0	22.5	31.5	48.7	23.5	33.0	48.5
	4.0	22.5	31.2	47.5	27.2	38.0	54.0	28.2	38.4	53.2
	3.0	26.2	36.0	51.0	31.2	40.7	57.5	32.2	43.1	57.2
	2.0	30.5	41.2	56.2	36.0	45.5	62.2	37.0	47.0	62.0
	1.5	35.0	45.2	60.2	38.7	49.0	65.7	40.0	51.0	65.0
	1.0	37.0	46.7	62.2	41.0	50.5	67.7	42.0	53.2	67.0
	0.0	179.5	401.2	653.7	180.5	398.2	651.7	180.0	403.7	658.0
CD P>0.01)			9.4	13.2	8.8	12.7	7.2	15.4	12.6	14.0

A study on the antifeedant activities of the leaf oils of *Callistemon lanceolatus*, *Cymbopogan winterianus*, *Eucalyptus sp.*, *Lantana camara*, *Nerium oleander*, *Ocimum basilicum*, *O. sanctum* and *Vitex negundo* was conducted against *Spodoptera litura* third instar larvae. The essential oils were applied at 2.5 and 10 % concentrations in the antifeedant assay. At 10% *O. sanctum*, *O.basalicum*, *C.winterianus*, *C. lanceolatus* and *V. negundo* caused 100% deterrence of feeding (Sharma *et al.*, 2001).

PROTEIN OR CUTICULIN INHIBITORS

Proteins form an integral part of the cuticle and play an important role in melanisation and sclerotisation. While great majorities of proteins are basic, the hydrophilic proteins present in larval cuticle of Lepidoptera have only relatively week interactions permitting the stretching and expansion during larval maturation (Anderson *et al.*, 1986, 1995). There has been increasing evidence on the effect of insect growth regulators (IGR) on cuticular protein synthesis. Juvenile hormone and its analogs alter the protein content and retain cuticle characteristic juvenile protein profile. Research on the influence of botanicals as protien or cuticulin inhibitors on insects is very little.

Lepidopterans treated with Azadiractin and plumbagin showed disturbance in cuticulogenesis visible externally as unsclerotized incompletely tanned areas in pupai state (Mordue and Black well, 1993; Krishnayya and Rao, 1995)

Analysis of cuticular protein was carried out to ascertain the effect of Plumbagin and azadirachtin on cuticulogenesis at larval-pupal transformation (Kumar and Subrahmanyam, 2000). Urea soluble cuticle protein content of the control insects reached a maximum at the late pupal stage and the treatment reduced the protein content significantly. The cuticular protein profile of the treated insects showed variation in the number and prominence of bands compared to that of control insects.

GROWTH REGULATORS

The two main groups of insect growth regulators (IGRs) being used on a commercial scale are the juvenile hormone analogue (JHAs) and the chitin synthesis inhibitors (CSIs). The JHAs have been tested in large-scale trials for the control of various pests of agriculture, forestry and public health importance. Chitin synthesis inhibitors belonging to the acyl urea group are also being used commercially for the control of a number of foliage feeders and tissue borrers. The role of botanicals as insect growth regulators has been evaluated to an extent. Insect growth regulating factors as antijuvenile hormonal agents have been reported from the extracts of the common goat weed *Ageratum conyzoides* Linn (Fagoonee and Umrit, 1980). There is a report from Jilani *et al.* (1984) stating sweat flag oil to have growth inhibitory properties against red flour beetle.

Studies carried out to evaluate plant extracts for their growth inhibitory effects on *Amrasca bigutula bigutula* (Patel and Patel, 1996) revealed that spraying of Repelin , Margocide and Neemark at 1% and Ardusa at 5% leaf suspensions and neem seed kernel suspension also at 5% level on bhendi showed a growth inhibitory effect on the pest and resulted in a lower percentage of normal adults.

An evaluation of the insecticidal growth inhibitory activity of essential oils of some medicinal plants was taken up by Sharma *et al.* (2001). Growth inhibitory activity of the leaf essential oils of *Callistemon lanceolatus*, *Cymbopogan winterianus*, *Eucalyptus sp.*, *Lantana camara*, *Nerium oleander*, *Ocimum basilicum*, *O. sanctum* and *Vitex negundo* was conducted against *Spodoptera litura* third instar larvae. The essential oils were applied at 2.0, 1.0, 0.5 and 0.025% in the insect development assay. Among these species, only *O. sanctum* caused mortality (20%). After 4 days exposure of the larvae to *O. sanctum* treated food, larval weight gain was lower than in the control.

Root extract of *Acorus calamus* at 1% conc. significantly reduced the larval survival and weight after ten days feeding and 1% of the extract of *Rauvolffia serpentina* resulted in 100% mortality of the larval population after 10 days feeding.

OVICIDAL AND OVIPOSITIONAL DETERENCY

The ovicidal effect of extracts of common bedding plant A.houstonianum Mill has been reported on large milkweed bug *Oncopeltus fasciatus* (Bowers *et al.*, 1976). The extract of *A. conyzoides* has also been found to inhibit the egg hatching as well as ovarian development in *D. flavidus*. Sign (Fagoonee and Umrit, 1980). Saxena *et al.* (1981) reported that, the dipping of eggs of rice leaf folder *Cnephalocrosis medianalis* Guenee into neem oil prevented the emergence of first instar larvae.

Azadirachtin was tested against a number of insects and was shown to be a strong oviposition deterrent (Warthen, 1989)

The oviposition deterrence and ovicidal properties of powders made from 15 plant species against *Callosobruchus maculatus* in stored cowpea seeds were tested. (Ofuya 1990). Fewest eggs were laid on seeds mixed with *Nicotiana tabacum* powder. Few eggs were also laid on seeds mixed with powders obtained from the bark of *Erythrophleum suaveolens* and the shoot of *Ocimum gratissimum*. Tobacco powder was the most effective in reducing the egg hatch followed by *E. suaveolens* powder.

The extracts from the seeds of the neem tree *Azadirachta indica* are known to posses fecundity reducing properties for various insects (Schmutterer, 1990).

Feeding and oviposition deterrent activities of an aqueous extract of *Tanacetum vulgare* in two of the cabbage pests, DBM and *Pieris rapae* was reported by Goldstein and Hahn (1992). Again, Hermawan *et al.* (1994) suggested that crude extract from *Andrographis paniculata* suppress oviposition by DBM females. The deterrent effects of Chinaberry fruit *Melia azaridirach* on oviposition of the DBM were reported by Dilwari *et al.* (1994) and Chen *et al.* (1996)

The effect of four botanicals on the oviposition of *Callosobruchus maculatus* F.was assayed by Rajapakse (1996). The plants selected for the study were *Piper nigrum, Anona reticulata, Dillenia retusa* and *Ocimum sanctum* in powder form. The powder obtained from fruits of *P.nigrum* significantly reduced oviposition and adult emergence while 100% mortality was observed at a higher conc. of 42%. The powder did not show any fumigant effect. Volatile oil obtained at 0.8% and above completely suppressed oviposition. The other plants tested did not significantly effect oviposition

Nair (1996) judged the ovicidal action of Acorus calamus extracts on melon fly Bactrocera cucurbitae by exposing the eggs to A.calamus extracts and observing for the egg mortality. As evidenced by the hatching percentage, the hatchability of the treated eggs were observed and egg mortality were recorded. Aqueous extract did not show ovicidal action however solvent extract showed as evidenced in the table 5.

Table 5. Comparison of size of female reproductives of normal and treated *B.cucurbitae*

	Normal ovary (20th day)		Treated ovary (20th day)		Normal-ovary (5th day)	
	Length (μ)	Breadth (μ)	Length (μ)	Breadth (μ)	Length (μ)	Breadth (μ)
R1	1658.25	1363.45	479.05	368.50	589.60	552.75
	1695.10	1326.60	442.20	368.50	626.40	663.30
R2	1621.40	1289.75	405.35	405.35	552.74	552.75
	1658.25	1289.75	442.20	442.20	552.75	589.60
R3	1695.10	1363.45	515.90	405.35	589.60	552.75
	1731.95	1289.75	479.05	442.20	626.45	552.75
R4	1768.80	1289.75	405.35	368.50	663.30	579.90
	1768.80	1400.30	442.20	405.35	626.45	589.60
R5	1768.80	1289.75	442.20	442.20	626.45	626.45
	1695.10	1363.45	515.90	368.50	626.45	663.30
Average	1706.15	1326.60	456.94	401.66	608.02	589.91

0.01%methanol extract of *A.calamus*

The size of ovaries of normal mature female flies and treated flies treated with 0.1%methanol extract from the day of emergence were dissected on the 20[th] day after emergence. The size of normal immature ovary dissected five days after emergence is also presented here for comparison (Table 6).

Table 6. Mean percentage mortality of eggs of *B. cucurbitae* treated with graded doses of *A.calamus* extracts

Treatment	Dose (%)	Mean % mortality*
Aqueous extract	0.10	0
	0.50	0
	1.00	0
	5.00	0
	10.00	0
	Control	0
Solvent extract	0.08	96.67
	0.06	90.00
	0.04	70.00
	0.02	36.64
	0.01	6.67
	Control	0.00

*Mortality observed after 24 hours

Bhargava *et al.* (1997) reported that alkaloid extracted from *Adhatoda vasica* could be effectively used against *Rhesala imparata* Wlk. and reported that deterred ovoposition of Tea mosquito bug

Adult pairs of males and females of *Earius vittella* Fab.a pest of cotton and okra were released to evaluate the effect of odours of the leaves of neem, lantana, eucalyptus tulsi and bulbs of garlic and one control set for their oviposition deterrent activity. All treatements significantly reduced the egg output as compared to the control. 9172 eggs). Similarly, all the odours significantly reduced the egg ha5tching compared to the control. (90.8%). The lowest number of eggs (128) and hatching (68.15%) were recorded with neem leaves odour.

The effects of extracts of *Leucas aspera, Ocimum sanctum, Azadiracta indica, Allium sativum* and *Curcuma longa* against the mosquito for their ovipositional deterrency was assayed by Murugan and Jayapalan (1999).

Ovipositional deterrency was significantly greater in *A.indica*, *L.aspera*, and *O. sanctum*.

The oviposition deterrent and toxic effects of seven plant sp. With known biological effects were tested against *Callosobruchus chinensis* L. on stored been seeds for two years at Ethiopia (Mulatu and Gebremedhin 2000). Oils of *A. indica*, *Milletiaie ferruginea* and *C. cineraraefolium* were the most effective in partially or completely preventing the egg laying and no bruchids emerged from the eggs laid.

In a detailed study to estimate the effect of extracts from *Clerodendron siphonanthus* leaf, a fence plant, the percentage of antifertility on the stored grain pest *Callosobruchus chinensis* (L), by treating through dipping methods was adopted (Pandey and Khan 2000). The three dosages tried were 0.5, 1.0 and 2.5 obtained by diluting the ethyl alcohol extract of the leaves. The newly emerged females were then released with untreated males into cages. After 20 to 25 days, eggs laid by each female were examined and viable eggs separated from the unviable eggs. The maximum percentage of reproduction control i.e. 88.4% was achieved with a 2.5% dose.

	Before Spraying	*After Spraying*
1. ***REPELLENT*** *Prevents pest influx.*		
2. ***OVIPOSITION DETERRENT*** *Deters egg laying*		
3. ***OVICIDE*** *Prevents & kills egg embryo*		
4. ***ANTIFEEDENT*** *Stems feeding on crops*		
5. ***INSECT GROWTH INHIBITOR*** *Blocks growth hormones and inhibits insect growth.*		

CONCLUSION

So far some of the major botanicals used in the pest management which are naturally occurring and have no adverse environmental impacts. The unilateral approach in pest control using chemical insecticides alone have an array of problems like resurgence and pesticidal resistance. Above all accumulation of toxic residues in the produce impairing health of the individual. Besides these problems, envronmentl problems also occurs. Here comes the importance of botanicals. It functions as antifeedant,ovipositional deterrant,cuticular protein inhibitor and also posses ovicidal action It can be included as a successful component ecofriendly management strategy.There are so many other plants which are under exploited and there is an immense scope for exploiting these botanicals in pest management.

DISCUSSION

Q.1 Among different species of Plumbago which one is most effectively used in the pest management strategy? Why?

Ans. Among plumbago, Plumbago rosea is most effective one, which is due to high content of plumbagin

Q.2 Whether Botanicals can be alone used as pest management strategy?

Ans. Botanicals alone cannot be used as a control measure it can be included as a tool in the pest management practice.

Q.3 Whether the neem is effective against tea mosquito bugs even though it attacks neem?

Ans. It is effective. There are some reports, which states that there will be reduction in the puncturing hole after the application of neem. This shows antifeedancy to the insect.

Q.4 Whether it is possible to apply neem in plantation crops?

Ans. It is possible. Neem is a botanical insecticide and also highly degradable in sunlight. So there, will be repeated application is required. So the application cost will be increased, that is the only disadvantage.

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DEVELOPMENT AND REGISTRATION OF NEW MOLECULES OF INSECTICIDES

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ABSTRACT

New technologies are available in line with the overall global development in pesticide science and technology. Substantial advances have been made in the area of newer pesticides and their technology. The advances are in line with the new cropping patterns. Agricultural chemicals used in crop/ life systems should have higher efficacy, greater safety, and lesser impact on environment. It is very important that such products are properly identified and evaluated by rigorous testing. Synthesis and development of an absolutely new pesticide is very costly and time consuming. To bring out just one successful, commercially viable pesticide, usually an average of about 10,000 compounds are synthesized and screened with an expenditure of about 25-30 crore or even more and a total time of six to eight years (Sivasankaran, 1995). Others are discarded.

Presently 165 pesticides are registered under the insecticides Act 1968. In recent years, the consumption of pesticides has shown a downward trend from 75,000 Mt in 1990-91 to around 46,155 MT in 1999-2000.However, the installed production capacity of pesticides in the country is around 1,25,000 MT annually with more than 125 manufacturing units (technical grade) and over 800 formulation units (Diwakar, 2000).

Agrochemical research over the past decades has resulted in the discovery of chemically novel insecticides that modify insect behavior, growth and development. The new molecules namely, benzoyl urea, nicotinoids, spinosyns, antibiotics and pyrazoles are more selective and safer to man and beneficial organisms (Ware, 2001). Our defence against pest complex in tomorrow's world will be compounds that are economically attractive, ecologically safe and user friendly.

INTRODUCTION

Agricultural chemicals used in crop/ life systems must be acceptable, environmentally safe and least hazardous to non-target species including man and domestic animals. It is very important that such products are properly identified and evaluated by rigorous testing. This is particularly with the toxic pesticides. Though the efforts are made to minimize the use of chemical pesticides by implementing IPM technology in our country, they still continue to dominate plant protection scenario.

The Kerala folidol tragedy during late fifties had taken toll of over 100 lives in Kerala and Tamil Nadu due to consumption of wheat smeared with folidol (ethyl parathion) due to leakage of containers during shipment. Takkar commission enquired the tragedy. The commission recommended to the GOI to bring out a comprehensive legislation on pesticides for their compulsory registration before their manufacture, transport, import/export, storage and use could be permitted

Presently 165 pesticides are registered under the Insecticides Act 1968. In the recent years, the consumption of pesticides has shown a downward trend from 75,000 Mt in 1990-91 to around 46,155 MT in 1999-2000.However, the installed production capacity of pesticides in the country is around 1,25,000 MT annually with more than 125 manufacturing units (technical grade) and over 400 formulation units. In the recent years, export of pesticides has gone up sharply. The average per hectare consumption of pesticides in India is around 30 mg. (Diwakar, 2000)

In the process of implementation of Insecticides Act 1968, 21 pesticides have been banned for use in the country (Aldrin, Benzenehexachloride (BHC), Calcium cyanide, Chlordane, Copper acetoarsenite, Dibromochloropropane (DBCP), Endrin, Ethylmercurychloride, Ethylparathion, Heptachlor, Menazon, Nicotine sulphate, Nitrofen, Paraquatemethylsulphate, Pentachloronitrobenzene (PCNB), Pentachlorophenol, Phenylmercuryacetate (PMA), Sodium Methane Arsonate (MSMA), Tetradifon, Toxaphene, Methomyl (24%)). The use of 10 pesticides is restricted and their use is allowed under strict supervision of trained officials. These pesticides are Aluminium phosphide, Captafol, Chlorbenzilate, DDT, Dieldrin, Ethylenebromide(EDB), Lindane, Methyl bromide, Methyl parathion and Sodium cyanide. In addition, so far 18 highly toxic pesticides have been refused registration for their use and manufacture in India.

DEVELOPMENT OF NEW INSECTICIDE MOLECULES

Synthesis and development of an absolutely new pesticide is very costly and time consuming. To bring out successful, commercially viable pesticide, an average of about 10,000 compounds have to be synthesized and screened with an expenditure of about 25-30 crore or even more and a total time of six to eight years. Most of the work on newer pesticides and technology development has been carried out in the USA, UK, France and other European countries, Japan and other technologically advanced countries.

Phase I- The process of adopting new pesticide evolved in other countries (taking into account the constraints of resources)

Phase II- Synthesis and screening of newer pesticide molecule

Phase I: Two basic steps involved in the introduction of a new pesticide in the market.

The selection of candidate pesticide

Its development and commercialization

Total time taken for this purpose is five to six years and the expenditure is Rs. 3 to 4 Crore

SELECTION

Selection of a new pesticide needs complete literature search and a lot of evaluation with respect to its efficacy and safety. There are a set of guidelines prescribed for such evaluations under the Insecticides Act, 1968.

DEVELOPMENT

Development of the selected pesticide for commercialization involves many stages in which the industry's R&D interacts with its own project engineering group, production staff, government agencies and marketing people.

The stages involved in pesticide development are :

1. Laboratory studies
2. Bench-scale and pilot plant studies
3. Data generation on bio-efficacy, residue(soil, water, crop) and toxicology(acute, sub acute and chronic)
4. Registration
5. Letter of intent (LOI) and satisfying letter of intent requirements
6. Industrial license, clearance from Pollution Board.
7. Plant installation, commissioning, and production
8. Marketing.

Minimum Requirement / Facilities to enable the company to launch the pesticide successfully

1.Library:

Literature pertaining to organic chemistry, entomology, pathology, agronomy, soil science, residue chemistry, toxicology and other aspects essential in the Library

2.Chemical Synthesis

Synthesis and process development for a particular identified compound is the first step. The process of manufacture has to be standardized with the locally available raw materials and the condition existing in the region. Well equipped Chemistry Laboratory with a batch of qualified and experienced chemists (synthetic and analytical) and chemical engineers. Normal requirements of equipments and facilities needed are well-established set of reaction kettles, cooling and vacuum systems, stirrers, High pressure and high temperature reactors, a pilot plant and a series of sophisticated instruments such as GLC, HPLC, IR, NMR and mass spectrophotometer

3.Bio-efficacy screening

Once the compounds are synthesized, they are to be screened against the target not only in the laboratory, but in the field as well, for establishing their effectiveness. In view of the variation in performance of pesticide under different climatic conditions, field evaluation for bio efficacy has to be carried out under various agro climatic regions. Facilities normally required for bio efficacy screening include the controlled environmental chamber, aseptic transfer desks, incubators, sprayers, micro applicators, laminar air flow tables and a well designed green house.

The role of the bio efficacy screening section does not end with the establishment of the relative performance of a compound. This section also has the responsibility of studying aspects such as (1) Field dosage determination (2) Phytotoxicity (3) Compatibility and monitoring of resistance development of a particular pest. The Laboratory should not only be equipped for such studies but also have qualified and experienced scientists in the field of specialization.

4.Residues and persistence

The manufacturer should ensure that proper and sufficient instructions are made available to the user to minimize the residue problems in food. To do this job, the manufacturer has to carry out extensive studies on the persistence and residues of each formulation. The facilities needed include well established laboratory with stirrers, blenders, evaporators, chromatographic systems and pecialized instruments such as GC, HPLC and GC- mass spectrometer. Even in these instruments, specialized detectors are needed for residue work in order to achieve high sensitivity. Electron capture, thermionic and flame-photometric detectors in case of GC and fluorescence detector of HPLC.

5.Toxicology

The most important item of pesticide safety evaluation is based on the toxicity to mammals, birds, fish and beneficial arthropods. Mammals: Acute, sub acute, chronic and field exposure with different routes of exposure. Only 4-5 laboratories have facilities to generate complete data on sub acute, oral(rat and dog), dermal(rabbit) and inhalation toxicity.

As far as data generation on chronic and supplemental toxicology is concerned, there are four centers that have the facilities to carry them out. In addition to the actual feeding of animals, other observations on histopathology and biochemistry play a major role. Only a few scientists are available on this study (These studies need a highly specialized and special breed of scientist).In the toxicological work, the primary factor is use of the good laboratory practices(GLP) standard. Unless this is adopted, the results may not be compared with the data obtained else where.

The facilities for full-fledged toxicology data generation laboratory include (1) a large animal breeding programmer,(2) sterile experimental rooms with complete displacement type air-conditioning, fully automatic autoclaves, air curtains, UV lamps, and a host of instruments such as tissue processors, micro tome, microscopes, auto analyzers for blood and urine and liquid scintillation counter.

6) Documentation

Computerized documentation center is required for recording and documenting all the data pertaining to the study. All the records will be retained in the archives for the period specified by the appropriate authorities. For any new insecticides to be a commercial success, not only its proper identification, development and cost effectiveness are important but also a strong and dedicated R& D work force to provide the necessary solutions whenever problems are encountered during manufacture, application, storage and handling. Thus an integrated approach is necessary in the launching of any pesticide. (Sivasankaran,1995)

CENTRES WHICH HAVE BETTER LABORATORY FACILITIES

Government

Central Insecticides Laboratory, Faridabad.

Industrial Toxicology Research Centre, Lucknow.

National Institute of Occupational Health, Ahmedabad.

Haffkine Institute, Bombay

Central Food Technological Institute, Mysore.

Private

Shri Ram Institute of Industrial Research, New Delhi.

Indian Institute of Toxicology, Bombay.

Rallis Agrochemical Research Station, Bangalore.

Sarabhai Research Centre, Vadodara.

Fredrick Institute of PP and Toxicology, Padappai.

Pesticides Research Laboratory, Calcutta.

Jai Rsearch Foundation, Vapi.

REGISTRATION OF NEW MOLECULES OF INSECTICIDES

A. Registration of Synthetic Insecticides
B. Registration of Plant Products
C. Registration of House hold Insecticides
D. Registration of *Bacillus thuringiensis*

1.REGISTRATION OF SYNTHETIC INSECTICIDES

Any person desiring to import or manufacture an insecticide has to apply for registration of the product to the Registration Committee set up under section 5 of the Act. The application is to be made in the form prescribed under rule 6 of the Insecticides rules 1971 and separate applications have to be submitted for each of the formulations and technical material. Each application has to accompany a registration fee of Rs100/- in the form of treasury challan / bank draft. Samples of labels, leaflets, and containers are also required to be submitted. The applicants are required to submit necessary scientific data in support of efficacy and safety of the product. On an application for registration the Registration Committee has to take a decision within a period of 18 months of the receipt of application. Guidelines on the data requirement for the grant of registration under the Insecticides Act 1968

Chemical composition and allied data requirements

1.Source of supply

The name and address of the supplier should be submitted for the registration of technical material for import.

2.Chemical composition

Complete chemical composition should be submitted both for the technical grade pesticides and their formulations. The minimum purity of the technical grade material should be clearly indicated and the details of the impurities should be submitted. Complete data on the identity and the physio chemical properties of the active ingredient should be submitted while applying for registration of Technical grade pesticides. The chemical composition for the formulations should include the kind, name and percentage of all the ingredients that are to be used in the formulation.

3. Specification

Complete Specifications for the product quality are to be submitted as per the ISI format. If ISI specifications are available, only the relevant IS number must be indicated.

4. Analytical Test Report

An Analytical Test Report giving the analysis of a particular batch for all the parameters included in the specifications should be submitted. The report should be duly signed by a qualified Chemist/Analyst.

5.Shelf-life data

The applicant should indicate the expected shelf-life claim for the product along with data in support of this claim.

6.Methods of analysis

Methods of analysis are to be submitted for those pesticides and their formulations for which the same are not available in ISI specifications.

Packaging and Labeling Requirements

No person shall stock or exhibit for sale or distribute any insecticide unless it is packed and labeled in accordance with the provision of these rules. Every package containing an insecticide, shall be of a type approved by the Registration Committee and a sample container in which the insecticide is proposed to be packed, shall either accompany the application for registration or shall be supplied to the Registration Committee separately.

Leaflet to be contained in a package: The packing of every insecticide shall include a leaflet containing the following details:

The plant disease, insects or noxious animals or weeds for which the insecticide is applied, the adequate direction concerning the manner in which the insecticide is to be used at the time of application. Particulars regarding chemicals harmful to human being, animals and wild lives, warning and cautionary statements including the symptoms of poisoning, suitable and adequate safety measures and emergency first aid treatment where necessary.

Caution regarding storage and application of insecticides with suitable warnings relating to inflammable, explosive or other substances harmful to the skin. Instructions concerning the contamination or safe disposal of used containers. A statement showing the antidote for the poison shall be included in the leaflet and the label. If the insecticide is irritating to the skin, nose, throat and eyes, a statement shall be included to that effect.

Manner of Labeling

The following particulars shall be either printed or written in indelible ink in the label of the innermost container of any insecticide and on the outermost covering in which the container is packed:

a) Name of the manufacturer (if the manufacturer is not the person in whose name the insecticide is registered under the Act, the relationship between the person in whose name the insecticide has been registered and the person who manufactures, packs or distributes or sells shall be stated.

b) Name of the insecticide (brand name or trade mark under which the insecticide is sold)

c) Registration number, Kind and name of the active ingredients and percentage of each insecticide.

d) Net content of volume (the net content shall be exclusive of wrapper or other material. The correct statement of the net content in terms of weight, measure, number of units of activity, as the case shall be given. The weight and volume shall be expressed in the metric system)

e) Batch number, f) Expiry date, g) Antidote statement

h) The Label shall be so affixed to the container that it can not be ordinarily removed.

The Label shall contain in a prominent place and occupying not less than 1/16 th of the total area of the face of the label, a square, set at an angle of 45 Deg. The dimension of the said square shall depend on the size of the package on which the label is to be fixed. The said square will be divided into two equal triangles, the upper portion shall contain the symbol and signal word and lower portion shall contain the colour specified. The upper portion of the square shall contain the following symbols and warning statements

a) Insecticide belonging to category I (extremely toxic) shall contain the symbol of skull and cross bones and the word POISON printed in Red. The following warning statements shall also appear on the label at appropriate place, outside the triangle (i) "KEEP OUT OF THE REACH OF CHILDREN" and (ii)"IF SWALLOWED OR IF SYMPTOMS OF POISONING OCCUR, CALL PHYSICIAN IMMEDIATELY"

b) Insecticides in Category II (Highly toxic) will contain the word POISON printed in Red and the statement KEEP OUT OF CHILDREN shall also appear on the label at appropriate place outside the triangle.

c) Insecticide in category III (Moderately toxic) shall bear the word DANGER and the statement KEEP OUT OF CHILDREN shall also appear on the label at appropriate place outside the triangle.

d) Insecticide in Category IV (Slightly toxic) shall bear the word CAUTION

The lower portion of the square should contain the colour depending on the classification of the insecticide: Extremely toxic – bright red, Highly toxic - bright yellow, Moderately toxic-bright blue and Slightly toxic- bright green. In addition to the precaution to be undertaken as specified, the label to be affixed in the package containing insecticide which are highly inflammable shall indicate that it is inflammable.

The label and leaflet to be affixed or attached to the packages containing insecticide shall be printed in Hindi, English and in one or two regional languages of the areas where the said packages are likely to be stocked, sold or distributed. Labeling of insecticide must not bear any unwarranted claims for the safety of the products or its ingredients. This includes statement such as SAFE, NON POISONOUS, NON INJURIOUS OR HARMLESS with or without such qualified phrase as when used as directed.

Transport and Storage of Insecticide

Packages contain insecticide, offered for transport by rail shall be packed in accordance with the conditions specified in the tariff issued by the ministry of railways. No insecticide shall be transported and stored in such a way as to come into direct contact with food stuff or animal feeds If any insecticide found to have leaked out in storage or transport, it shall be the responsibility of the transport agency or storage owner to take such measure urgently to prevent poisonings and pollution of soil or water if any.

The packages containing insecticides shall be stored in separate rooms or premises away from the rooms or premises used for storing other articles shall be kept in separate almera under lock and key depending upon the quantity and nature of insecticide. The rooms or premises meant for storing insecticide shall be well built, dry, well lit and ventilated and of sufficient dimension

Data Requirement on Bio efficacy and Residues

A. Experimental data on Bioefficacy

Bioefficacy test on the proposed formulation must be conducted under Indian condition in the laboratory and field. The tests are effectiveness of insecticides, phytoxicity, translocation, persistence in soil and water and plant, compatibility with other chemicals, direction and concerning the dosage and time of application, type of application equipment and the manner in which the insecticide is to be used

B. Experimental data on residue

The residue test must be conducted under Indian condition. The test includes residue analysis in food and feeding stuff, water and soil, expected residue level in edible crops and soil and worker hazard after treatment.

Data requirement on safety/toxicology

1. Basic Data includes registrant, name of the pesticide with % of the AI, brand name, common name and manufacturer name and address

2. Acute toxicity studies in mammals

Oral : rat and mice., Dermal : Rabbit and rat., Inhalation : any one species., Primary skin irritation: Rabbit., Irritation mucus membrane: any one species

3. Information on the following

Toxicity to pigeon and chicken, fresh water fish and bees.

4. Observation on man

Direct field observation on the population in case the technical material is used as such in the field

Health records on Industrial workers

5. Medical data

Record of poisoning cases, Science of poisoning, Diagnosis of poisoning and chemical test

Treatment of poisoning, First aid measure and Medical treatment

B. REGISTRATION OF PLANT PRODUCTS

Data requirement for registration of plant products:

1 Chemistry

Name of the parts of the plant to be used for the extraction and method of experiment in detail clearly identifying the chemical should be provided along with application form. Neem extracts contains azadirachtin as one of the major active ingredient, the concentration in the formulated neem extract should contain not less than 1500 ppm of azadirachtin in Kernel based formulation and 300 ppm in neem oil based formulation. When the insecticidal active ingredient is other than azadirachtin, then the applicant has to indicate the name, quality and quantity of that particular active ingredient. Other specification needed are physico Chemical Properties and specification of ingredients and analytical test report and Shelf life

2. Bioefficacy

Bio effectiveness, Phytotoxicity, Compatibilty with other chemicals, Direction concerning dosage

Time of application, Waiting period, Application equipment and Information regarding status in other countries if any

3. Toxicology

Acute Oral : Rat and mice., Acute dermal: Rat and mice

Primary skin irritation and irritation to mucus membrane, Pneuro behavioural toxicity, Reproductive Toxicity, Carcinogenecity, Mutagenecity and Effect of spray operator

4. Packaging and labeling

Labels and leaflets as per Insecticide rule and existing norms, Type of packaging, Manner of packaging containers content compatibility, Specification for primary package, Specification for secondary package, Specification for transport package, Manner of labeling, Instruction for storage and use, Information regarding disposal of used package and Process of manufacturing indicating material balance generation of waste.

C. REGISTRATION OF HOUSEHOLD PESTICIDE

Household insecticides are defined as a insecticides which are used for direct domestic application in the form of different devices in and around areas of all structures including vehicles, areas associated with the household of human life, patience care areas of health, related institutions or areas where children spent their time as in schools, garden etc in order to kill/repel the insects and vector of the disease

Classification

The committee classified the household insecticide into four categories depending on their physical chemical properties usages packaging requirements, method of application and data requirement.

Category I

The household Insecticides which are used as solid for the purpose defined under the definition will come under this category e.g baits and glue

Category II

The household pesticides which are used in the form of liquids for the purpose enumerated under definition will come under this category e.g ready to use spray, space and spot application, EC formulation and aerosols.

Category III

The household pesticides in technical form which emits vapours/gas/smoke eitheras such or as a result of heating, burning or coming in contact with air/moisture or with the help of any other appliances/technology e.g. mats, coils, cake etc

Category IV

The products which do not come under the above mentioned a) to c) categories but used for the purposes as mentioned in the definition will cover under this category.

Data requirement for registration of house hold insecticides include:

Chemical composition and allied data requirements, specification, analytical test report, shelf life data and method of analysis. Data on residue, bio efficacy and toxicology are same as other synthetic insecticides. Glass bottles as primary packaging material is not permitted for household pesticides.

D.REGISTRATION OF BIOCONTROL AGENTS

Data requirement for registration of bio control agents

1 Chemistry

Physico chemical specification and composition, systematic name and strain, common name, natural occurrence of the organism, manufacturing process and an appropriate test procedure and criteria used for identification should be provided in the application form. Method of analysis includes the protein content per mg, field test, data on persistence, data on residues, compatibility, time of application and data on non targeted organisms

2 Toxicity

Single Exposure Studies include oral toxicity, dermal toxicity, mucus membrane irritation, Inhalation toxicity and Alergy / Sensitisation / Immuno suppression

Repeated exposure studies are oral toxicity, dermal toxicity and Inhalation toxicity/Pathogenicity

Supplementary toxicity studies include mutagenicity, teratogenicity and Carcinogenicity

Eco-toxicity tests are toxicity to birds, fish, honey bee and silk worm

3 Processing, packaging and labeling

Manufacturing process/Process of formulation and information on raw material/Material balance, plant and machinery, process – Unit operation and finished products and generation of waste

Glass bottles are not recommended for bio control agents package.

PROCEDURE FOR ACCEPTANCE OF PESTICIDE TO BE APPLIED FROM AIR

1.Application

The application may be made to the plant protection advisor to the Government of India, Ministry of agriculture, Directorate of plant protection and storage, Faridabad, Haryana.

2.Definitions

"ULV" means quantity less than one gallon per acre

"From air means" application through an aerial spraying equipment fitted to an aircraft or a helicopter

"Pesticide" includes insecticide, fungicide, herbicide, acaricide and rodenticide

3. Information/Data required are registration number and following scientific data may be supplied with the application, accepted common name of pesticide, chemical formula, formulation with constituents, physical properties of the formulation, specific gravity, volatility, viscosity and flash point

4 Chemical properties of the formulation to be attached are solubility, stability, odour and colour

5 Safety data include acute mammalian studies : Oral and Dermal, Inhalation, eye and skin irritation and short term studies in Dogs for a duration of 90 days. Toxicity to fish, bees, live stock and parasite and predators.(David,1992)

NEW MOLECULES IN PEST MANAGEMENT

Conventional pesticides such as chlorinated hydrocarbons; organo phosphates, carbamates and pyrethroids were successful in controlling insect pests during the past five decades, thereby minimizing losses in agricultural yields. Unfortunately, many of these chemicals are harmful to man and beneficial organisms and cause ecological disturbances. Also, the repeated use of conventional recommended insecticides posed problem such as resistance to insecticides, secondary pests outbreaks and resurgence of insect pests (Patil *et al.*,1990). Development of resistance to conventional insectcidal groups such as organophosphates, carbamates and endosulfan was detected in field populations of *H. armigera* in India. (Armes *et al.*,1996). This provides impetus to study new alternatives and more ecologically acceptable methods of insect control such as biorational approaches of pest management involving insecticides which adversely affects or modify the biochemical or physiological processes of the target organisms and poses multistage toxicity against a single insect pest.

NICOTINOIDS

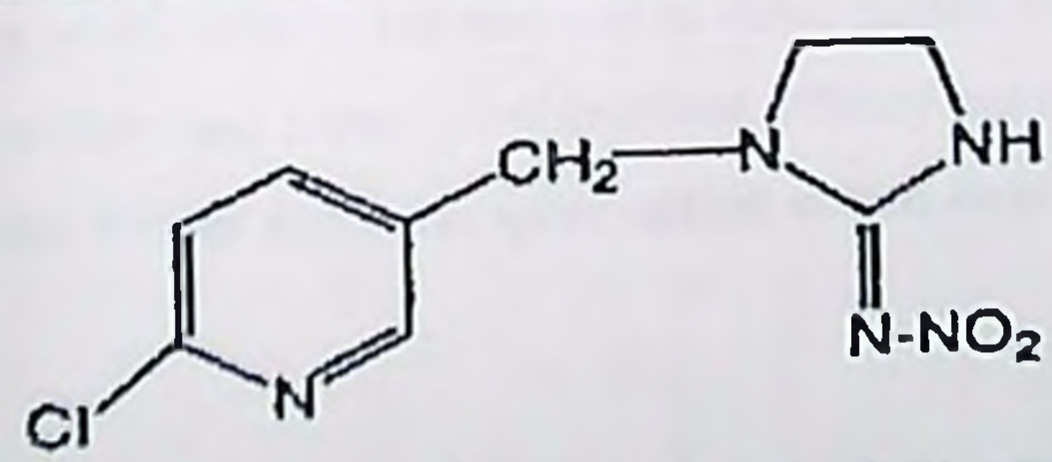
The nicotinoids are a new class of insecticides with a new mode of action. They have been previously referred to as *nitro-quanidines, neonicotinyls, neonicotinoids, chloronicotines,* and more recently as the *chloronicotinyls*. Just as the synthetic pyrethroids are similar to and modeled after the natural pyrethrins, so too, are the nicotinoids similar to and modeled after the natural nicotine.

Imidacloprid was introduced in Europe and Japan in 1990 and first registered in the U.S. in 1992. It is currently marketed as several proprietary products worldwide, e.g., Admire®, Confidor®,Gaucho®, Merit®, Premier®, Premise® and Provado®. Very possibly it is used in the greatest volume globally of all insecticides.(Ware,2001)

Imidacloprid is a systemic insecticide, having good root-systemic characteristics and notable contact and stomach action. It is used as a soil, seed or foliar treatment in cotton, rice cereals, peanuts, potatoes, vegetables, pome fruits, pecans and turf, for the control of sucking insects, soil insects, whiteflies, termites, turf insects and the Colorado potato beetle, with long residual control. Imidacloprid has no effect on mites or nematodes. Elbert *et al.*,(1991) reported that it has outstanding insecticidal activity against the sucking pest and has long persistent activity.

Other nicotinoids include acetamiprid (Mospilan®), thiamethoxam (Actara®, Platinum®), and nitenpyram (Bestguard®). All are pursuing U.S. registration.

IMIDACLOPRID (Gaucho®, Provado®)



1-(6-chloro-3-pyridin-3-ylmethyl)-N-nitro=
Imidazolidin-2-ylidenamine

Mode of action--The nicotinoids act on the central nervous system of insects, causing irreversible blockage of postsynaptic nicotinergic acetylcholine receptors

Cent per cent mortality of the aphids was observed up to seven and nine days after treatment in Imidacloprid 200 SL at 100 and 150 ml per hectare respectively. More than 50% mortality was observed.(Rameshbabu and Santharam, 2000)

Kumar (1998) reported that Imidacloprid 200SL foliar spray at 100 and 150 ml per hectare persisted for 22 days against *Aphis gossypii* and 30 days against *Amrasca devastans* in cotton.

Thiamethoxam – New Seed Dresser

It belongs to nitro guanidine group possessing potential activity against sucking pests and have a novel mode of action (Shinzo-kaajabu,1996). It is systemic insecticide. This has been visualized as an alternative over Soil and foliar applications of insecticides because of several factors such as low dose, less pollution, earliness, simplicity in application, selectivity and less interference in natural fauna. Its Efficacy was tested against sucking pest of cotton.

Results clearly showed that seed dressing with thiamethoxam at 4.3 g per kg of seeds keep the population of aphids, jassids and thrips below ETL Level that is no economic damage was caused by these pests up to 50 days . Moreover, it also suits in IPM strategy as all the seed dressing treatments encourages natural enemy population in cotton, besides controlling sucking pest. It enhance the growth (Phytotonic effect) of cotton plants (Vadodaria *et al.*, 2001)

Acetamiprid

It has a cyanodine structure which affects Ach receptor of insect central nervous system. It has high systemic and translaminar activity and hence gives excellent efficacy against sucking pest complex. It has been used both as seed dresser and foliar spray against cotton early sucking pests in irrigated condition.

Bioefficacy

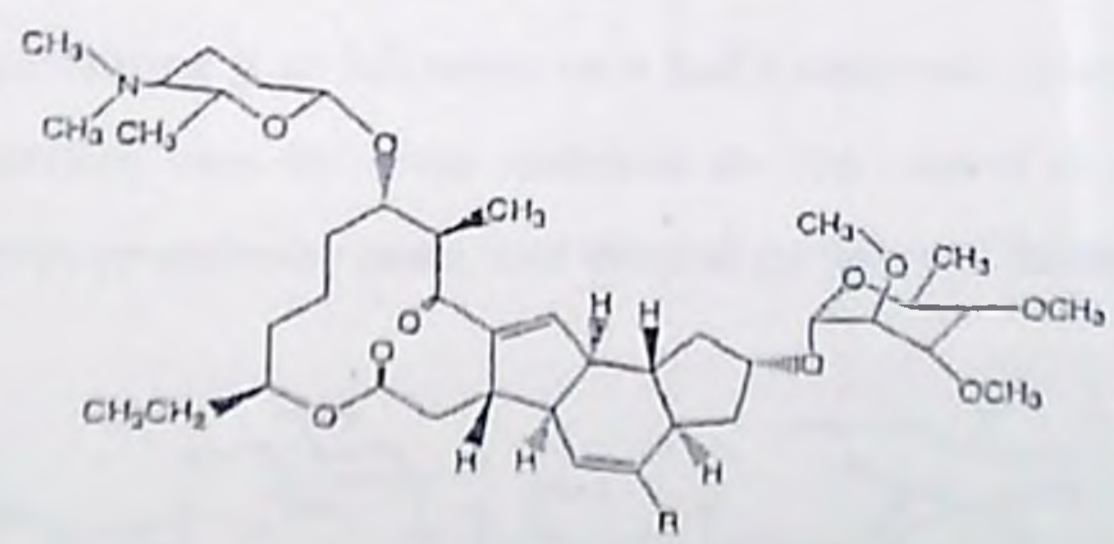
Results of the pooled analysis indicated that Acetamiprid 20SP at dosages 26.25 g/kg of seed protected the cotton crop up to 39 days against early sucking pests. Whereas two applications of Acetamiprid 20SP as foliar spray at 15 g a.i per hectare on ETL basis protected the crop up to 60 days effectively. Seed dressing has recorded higher population of predators (Patil *et al.*, 2001). This result was very much similar as reported by Kumar *et al*.,(1999). Acetamiprid 20SP also suits well in IPM Strategy as it effectively checks early sucking pest in irrigated cotton ego systems

SPINOSYNS

Spinosyns are perhaps the newest class of insecticides, represented by spinosad (Success®, Tracer Naturalyte®). Spinosad is a fermentation metabolite of the actinomycete *Saccharopolyspora spinosa*, a soil-inhabiting microorganism. It has a novel molecular structure and mode of action that provide excellent crop protection typically associated with synthetic insecticides, first registered for use on cotton in 1997. Spinosad is a mixture of spinosyns A and D (thus its name, spinosad).

It is particularly effective as a broad-spectrum material for most caterpillar pests at the astonishing rates of 0.04 to 0.09 pound of active ingredient (18 to 40 grams) per acre. It has both contact and stomach activity against lepidopteran larvae, leaf miners, thrips, and termites, with long residual activity. Crops registered include cotton, vegetables, tree fruits, ornamentals and others. It has low ma mammalian toxicity and selectively active on Lepidopteran pests (Adan *et al*., 1996).

Mode of action--Spinosad acts by disrupting binding of acetylcholine in nicotinic acetylcholine receptors at the postsynaptic cell. It also modifies receptor indirectly. It binds to one or more proteins and brings influx of sodium ions. Neurons get depolarized and muscles become hyperactive followed by neuromuscular fatigue, paralysis and death.



spinosyn A: R = H, MW = 731.98
spinosyn D: R = CH$_3$, MW = 746.00

Bio efficacy

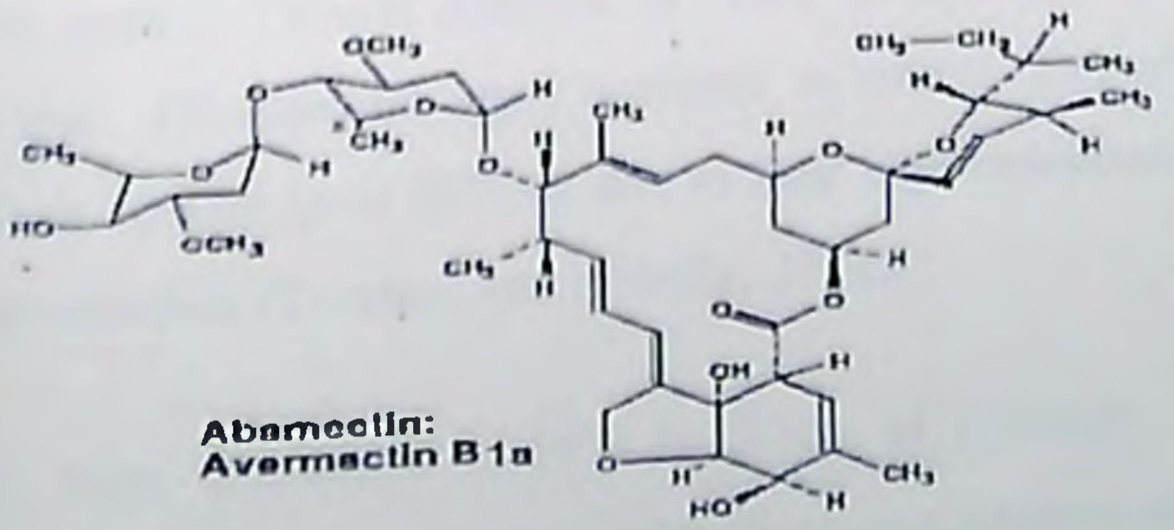
Spinosad 48 SC was evaluated against 3 most important lepidopteran pests of cabbage viz., diamond back moth *Plutella xylostella*, cabbage head borer *Hellula undalis* and cabbage leaf caterpillar *Spodoptera littoralis* at three seasons of 1995 to 1998 at different dose level from 10 to 25 g a.i per hectare. The experiment revealed that it provided effective control of all the three injurious lepidopterans and efficacy persisted for 7 days.

Spinosad @ 15 to 25 g a.i per hectare showed very little adverse effect on the two most important parasitoids (*Cotesia plutellae* and *Brachymeria sp*) of diamond back moth infesting cabbage in West Bengal (Dey and Somchowdhury ,2001) .

Spinosad has reported to be highly effective against number of lepidopteran caterpillars viz., *Hellula undalis*, *Crocidolomia binotalis* and *Plutella xylostella*. It has also been reported to be highly effective against *Helicoverpa armigera* on chickpea and on cotton. It is also reported to be safe to non target organisms viz., beneficial insect. Spinosad 48EC at 75g ai / ha recorded lower larval population and higher seed cotton yield (Vadodaria *et al.*, 2001).

ANTIBIOTICS

In this category belong the *avermectins*, which are insecticidal, acaricidal, and antihelminthic agents that have been isolated from the fermentation products of *Streptomyces avermitilis*, a member of the actinomycete family. *Abamectin* is the common name assigned to the avermectins, a mixture containing 80% avermectin B1*a* and 20% B1*b*, homologs that have about equal biological activity. Clinch® is a fire ant bait, and Avid® is applied as a miticide/insecticide. Abamectin has certain local systemic qualities, permitting it to kill mites on a leaf's underside when only the upper surface is treated. The most promising uses for these materials are the control of spider mites, leafminers and other difficult-to-control greenhouse pests, and internal parasites of domestic animals.



Emamectin benzoate (Proclaim®, Denim®) is an analog of abamectin, produced by the same fermentation system as abamectin. It is both a stomach and contact insecticide used primarily for control of caterpillars at the rate of 0.0075 to 0.015 lb (3.5 to 7.0 grams) a.i. per acre. Shortly after exposure, larvae stop feeding and become irreversibly paralyzed, dying in 3-4 days. Rapid photodegradation of both abamectin and emamectin occurs on the leaf surface.

Mode of action--Avermectins block the neurotransmitter gamma aminobutyric acid (GABA) at the neuromuscular junction in insects and mites. Visible activity, such as feeding and egg laying, stops shortly after exposure, though death may not occur for several days.

Bio efficacy

Vertimec 1.8EC @ 15 g a.i/ha was found to be highly effective in checking DBM larval population and also recorded significantly higher yield i.e 51-52 tonnes of marketable cabbages heads/ha while the untreated check recorded a yield of 33 tonnes/ha. Hence it can be concluded that the insecticide Vertimec 1.8EC is highly effective in reducing the level of incidence of DBM larval population on cabbage crop. This insecticide may be alternated with other effective insecticide to keep the DBM under manageable level (Murugan and Ramachandran ,2000).

BENZOYLUREAS

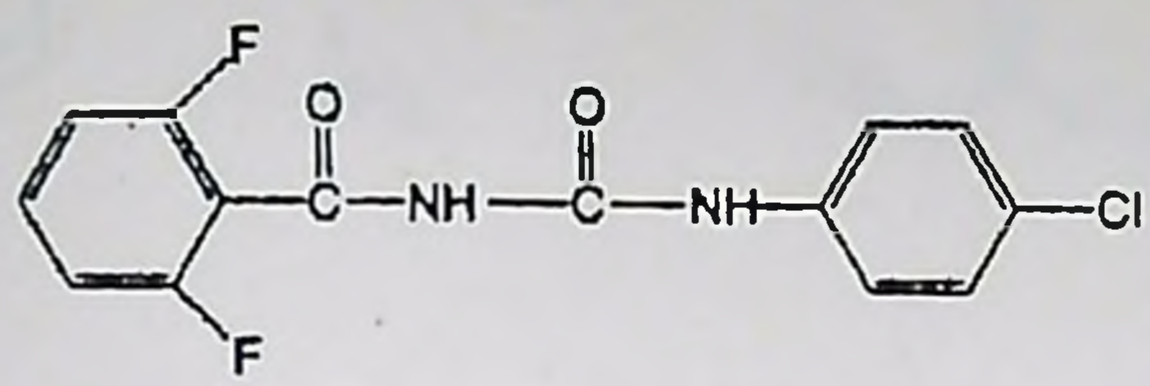
Benzoylureas are an entirely different class of insecticides that act as insect growth regulators (IGRs). Rather than being the typical poisons that attack the insect nervous system, they interfere with chitin synthesis and are taken up more by ingestion than by contact. Their greatest value is in the control of caterpillars and beetle larvae.

Benzoylureas were first used in Central America in 1985, to control a severe, resistant leafworm complex (*Spodoptera spp., Trichoplusia spp.*) outbreak in cotton. The withdrawal of the ovicide chlordimeform made their control quite difficult due to their high resistance to almost all insecticide classes, including the pyrethroids (Turnbull and Howells, 1982).

The first benzoylureas were introduced in 1978 by Bayer of Germany, triflumuron (Alsystin®) being the first. Others appearing since then are chlorfluazuron (Atabron®, Helix®), followed by teflubenzuron (Nomolt®, Dart®), hexaflumuron (Trueno®, Consult®), flufenoxuron (Cascade®), and flucycloxuron (Andalin®). Others are Flurazuron, Novaluron, and Diafenthiuron. Lufenuron (Axor®) is the newest addition to this group, appearing in 1990. Surprisingly, none of these are registered in the U.S.

The only benzoylurea registered in the U.S. is diflubenzuron (Dimilin®, Adept®, Micromite®). It was first registered in 1982 for gypsy moth, cotton boll weevil, most forest caterpillars, soybean caterpillars, and mushroom flies, but now with a much broader range of registrations.

DIFLUBENZURON (Dimilin®)



1-(4-chlorophenyl) 3-(2,6-difluorobenzoyl)urea

Though not a benzoylurea, cyromazine (Larvadex®, Trigard®), a triazine, is also a potent chitin synthesis inhibitor. It is selective toward Dipterous species and used for the control of leafminers in vegetable crops and ornamentals, and fed to poultry or sprayed to control flies in manure of broiler and egg producing operations, and incorporated into compost of mushroom houses for fungus gnats.

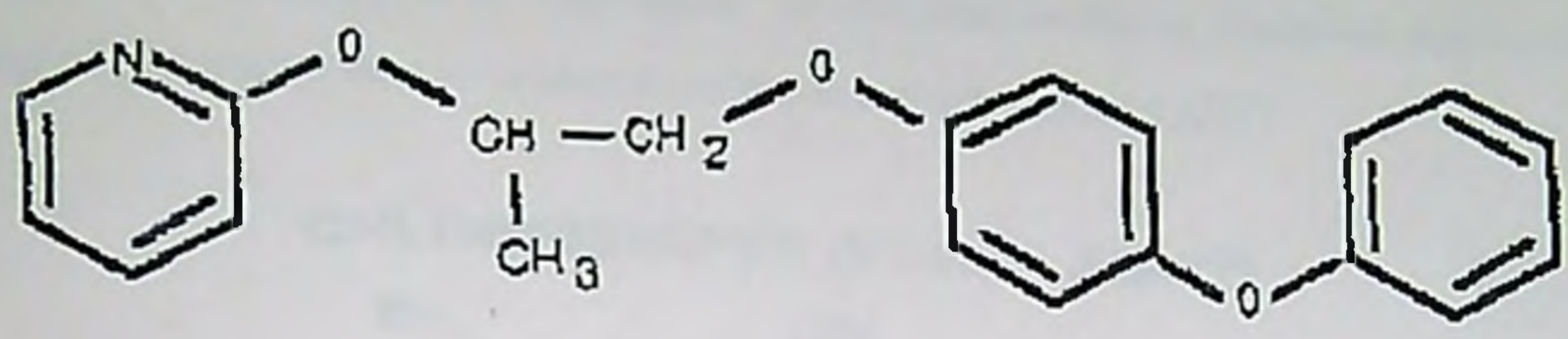
All the urea compounds exhibited very strong ovicidal action against eggs of all ages at 600 and 800 ppm (Chakraborti and Chatterjee, 2001). This high ovicidal action has been documented by Becker (1985) on *Plutella xylostella*., by Horowitz *et al* (1992) on *Earias vitella* and by Mani *et al* (1991) on *Sogatella furcifera*.

Post and Vincent (1973) reported that diflubenzuron interfered in the process of cuticle deposition and produced egg hatching inhibitory effect while Mulder and Gijwijt (1973) found that diflubenzuron caused failure in moulting and pupation leading to death by interfering in the process of cuticle deposition.

Mode of action--The benzoylureas act on the larval stages of most insects by inhibiting or blocking the synthesis of chitin, a vital and almost indestructible part of the insect exoskeleton. Typical effects on developing larvae are the rupture of malformed cuticle or death by starvation. Adult female boll weevils exposed to diflubenzuron lay eggs that do not hatch. And, mosquito larvae control can be achieved with as little as 1.0 gram of diflubenzuron per acre of surface water.

Buprofezin (Applaud®), classed as a thiadiazine IGR. Both have given excellent results in controlling the whitefly complex, now a universal problem in U. S. cotton production.

Buprofezin

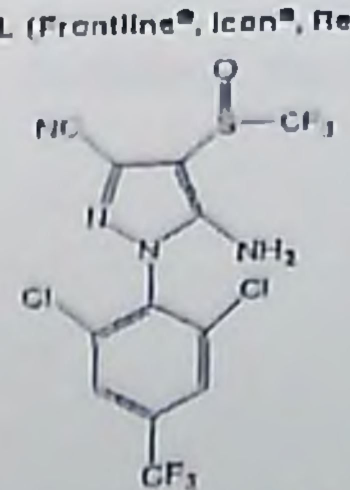


FIPROLES

Fipronil (Regent®, Icon®, Frontline®) is the only insecticide in this new class, introduced in 1990 and registered in the U.S. in 1996. It is a systemic material with contact and stomach activity. Fipronil is used for the control of many soil and foliar insects, (e.g., corn rootworm, Colorado potato beetle, and rice water weevil) on a variety of crops, primarily corn, turf, and for public health insect control. It is also used for seed treatment and formulated as baits for cockroaches, ants and termites. Fipronil is effective against insects resistant or tolerant to pyrethroid, organophosphate and carbamate insecticides.

Mode of action--Fipronil blocks the gamma-aminobutyric acid- (GABA) regulated chloride channel in neurons, thus antagonizing the "calming" effects of GABA

FIPRONIL (Frontline®, Icon®, Regent®)



5-amino-1-[2,6-dichloro-4-(trifluoromethyl)phenyl]-3-cyano-4-trifluoromethylsulphinylpyrazole

PYRROLES

Chlorfenapyr (Alert®, Pirate®) is the first and only member of this unique chemical group, as both a contact and stomach insecticide-miticide. It is used on cotton and experimentally on corn, soybeans, vegetables, tree and vine crops, and ornamentals to control whitefly, thrips, caterpillars, mites, leafminers, aphids, and Colorado potato beetle. It has ovicidal activity on some species.

Mode of action--Chlorfenapyr is an "uncoupler" or inhibitor oxidative phosphorylation, preventing the formation of the crucial energy molecule adenosine triphosphate (ATP)

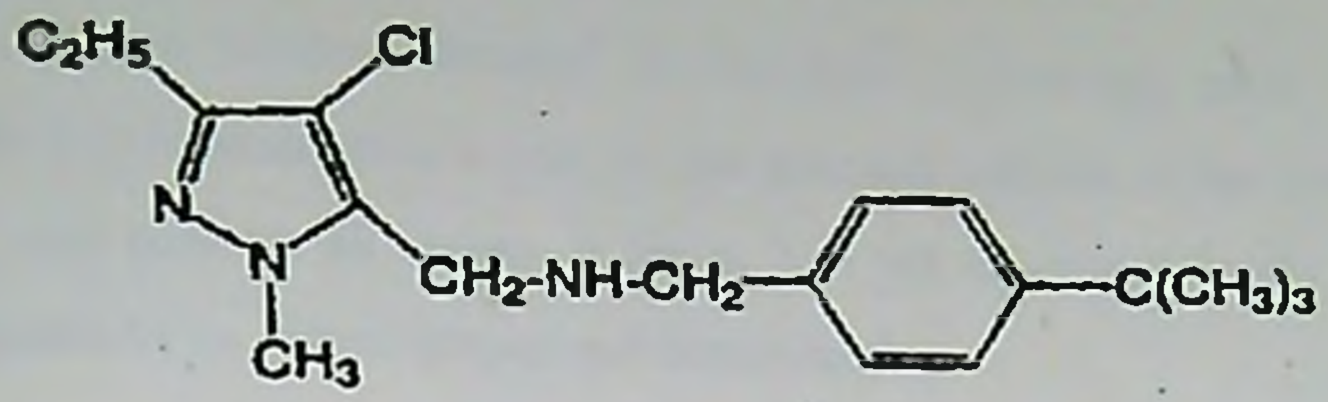
CHLORFENAPYR (Pirate®, Alert®)

4-bromo-2-(4-chlorophenyl)-1-ethoxymethyl-5-trifluoromethylpyrrole-3-carbonitrile

PYRAZOLES

The pyrazoles consist of tebufenpyrad and fenpyroximate. These were designed primarily as non-systemic contact and stomach miticides, but do have limited effectiveness on psylla, aphids, whitefly, and thrips. Tebufenpyrad (Pyranica®, Masai®) is used experimentally on cotton, soybeans, vegetables, pome fruits, grapes and citrus. Fenpyroximate (Acaban®, Dynamite®) controls all stages of mites, gives fast knockdown, inhibits molting of immature stages of mites, and has long residual activity. Fenpyroximate 5EC provide significant superior control of eriophyid mite *Aceria guerreronis* @ 10 ml/ palm in the case of root feeding and 0.75 –1.0 ml / liter of water as direct spraying and is much less toxic to predatory phytoseid mite *Amblyseius sp.* (Dey and Somchoudhury, 2001)

TEBUFENPYRAD (Pyranica®, Masai®)



**4-chloro-N[[4-(1,1-dimethylethyl)phenyl]methyl]-
3-ethyl-1-methyl-1H-pyrazole-5-carboxamide**

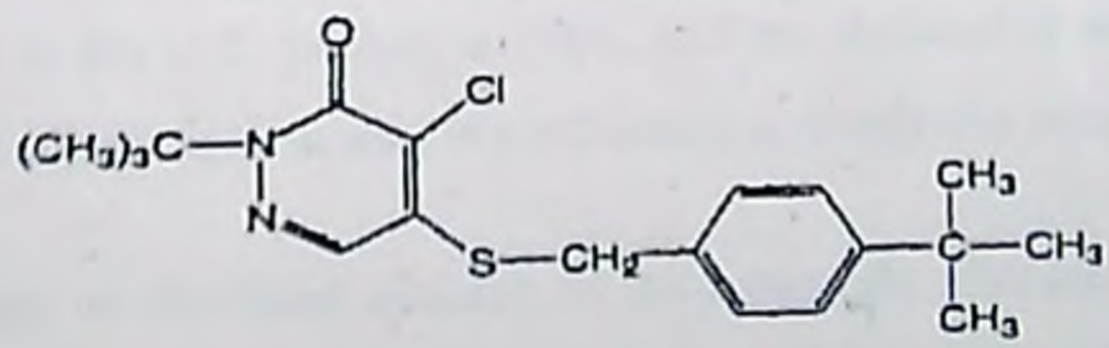
Mode of action--Their mode of action is that of inhibiting mitochondrial electron transport at the NADH-CoQ reductase site, leading to the disruption of adenosine triphosphate (ATP) formation, the crucial energy molecule.

PYRIDAZINONES

Pyridaben (Nexter®, Sanmite®) is the only member of this class. It is a selective contact insecticide and miticide, also effective against thrips, aphids, whiteflies and leaf hopprs. Registrations are for pome fruits, almonds, citrus, ornamentals and greenhouse ornamentals. Pyridaben provides exceptionally long residual control, and rapid knockdown at a broad range of temperatures.

Mode of action--Pyridaben is a metabolic inhibitor that interrupts mitochondrial electron transport.

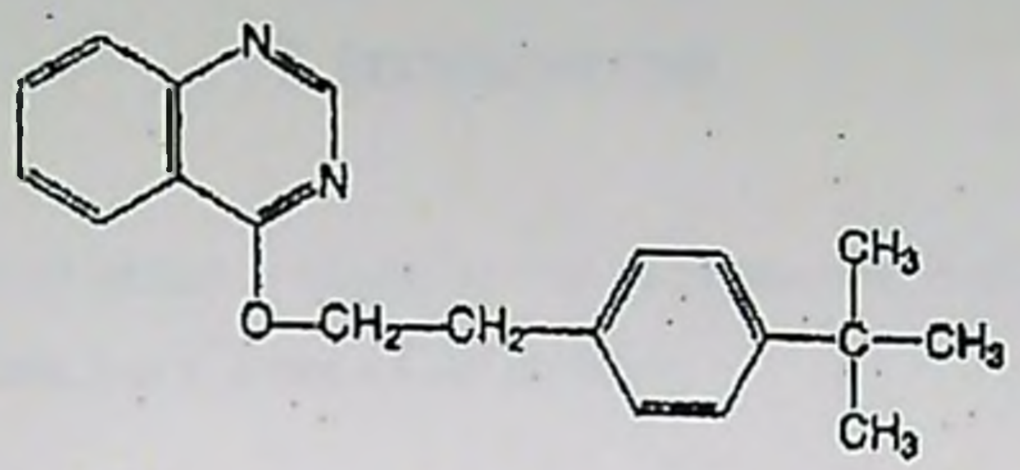
PYRIDABEN (Nexter®, Sanmite®)



**2-tert-butyl-5-(4-tert-butylbenzylthio)-4-
chloropyridazin-3(2H)-one**

QUINAZOLINES

The quinazolines offer a unique chemical configuration, consisting only of one insecticide, fenazaquin (Matador®). Fenazaquin is a contact and stomach miticide. It has ovicidal activity, gives rapid knockdown, and controls all stages of mites. Not yet registered in the U.S., it is used on cotton, stone and pome fruits, citrus, grapes and ornamentals.



4-[[4-(1,1dimethylethyl)phenyl]ethoxy]quinazoline

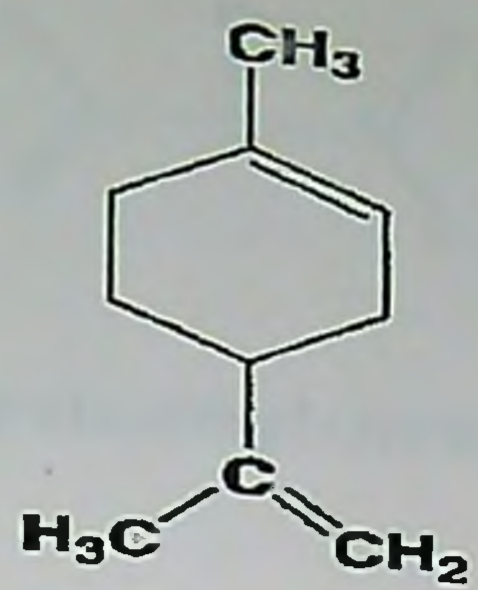
Mode of action--Fenazaquin inhibits mitochondrial electron transport at Site 1.

BOTANICALS

Botanical insecticides are of great interest to many, for they are natural insecticides, toxicants derived from plants. Historically, the plant materials have been in use longer than any other group, with the possible exception of sulfur. Tobacco, pyrethrum, derris, hellebore, quassia, camphor, and turpentine were some of the more important plant products in use before the organized search for insecticides began in the early 1940s.

Botanical insecticide use in the U.S. peaked in 1966, and has declined steadily since. Pyrethrum is now the only botanical of significance in use. We will mention briefly the others.

Limonene or *d*-Limonene is the latest addition to the botanicals. Extracted from citrus peel, it is effective against all external pests of pets, including fleas, lice, mites, and ticks, and is virtually nontoxic to warm-blooded animals. Several insecticidal substances occur in citrus oil, but the most mportant is limonene, which constitutes about 98% of the orange peel oil by weight.



Limonene

Mode of action--Its mode of action is similar to that of pyrethrum. It affects the sensory nerves of the peripheral nervous system, but it is not a ChE inhibitor.

Neem oil extracts are squeezed from the seeds of the neem tree and contain the active ingredient *azadirachtin*, a nortriterpenoid belonging to the lemonoids. Azadirachtin has shown some rather sensational insecticidal, fungicidal and bactericidal properties, including insect growth regulating qualities. Azatin®is marketed as an insect growth regulator, and Align®and Nemix®as a stomach/contact insecticide for greenhouse and ornamentals.

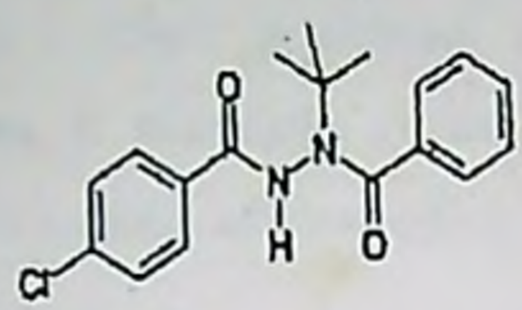
Mode of action--Azadirachtin disrupts molting by inhibiting biosynthesis or metabolism of ecdysone, the juvenile molting hormone.

INSECT REPELLENTS

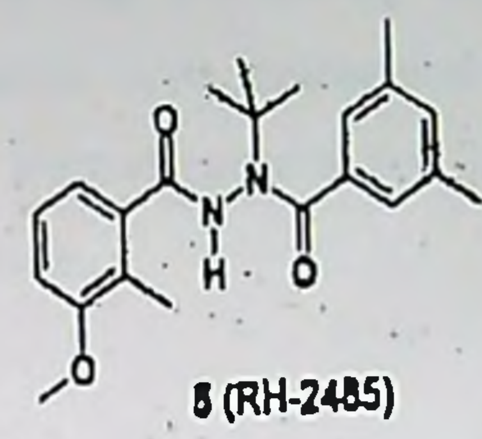
Historically, repellents have included smoke, plants hung in dwellings or rubbed on the skin as the fresh plant or its brews, oils, pitches, tars, and various earths applied to the body. Before a more edified approach to insect olfaction and behavior was developed, it was assumed that if a substance was repugnant to humans it would likewise be repellent to annoying insects.

In recent history, the repellents have been dimethyl phthalate, Indalone®, Rutgers 612®, Dibutyl phthalate, various MGK®repellents, Benzyl benzoate, the military clothing repellent (N-butyl acetanilide), Dimethyl carbamate (Dimelone®) and Diethyl toluamide (DEET, Delphene®). Of these, only DEET has survived, and is used worldwide for biting flies and mosquitos. Most of the others have lost their registrations and are no longer available.

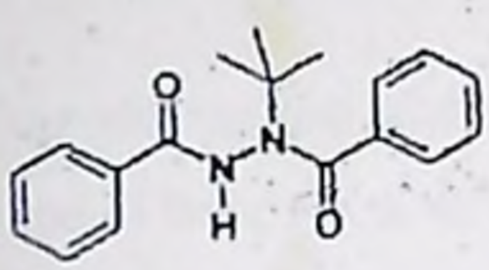
RH 2485 : (Methoxy fenozide)The newest member of bisacylhydrazine is under development by Rohm and Hass company. This compound is significantly more active than tebufenozide. It has wide range of activity against lepidopteran pests of cotton, corn and other major agronomic crops. It has no adverse effect on no target organisms



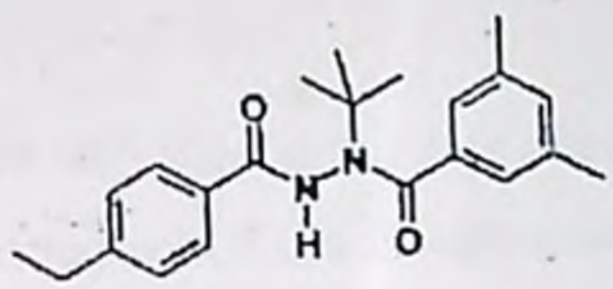
4 (RH-0345, halofenozide)



5 (RH-2485)



2 (RH-5849)

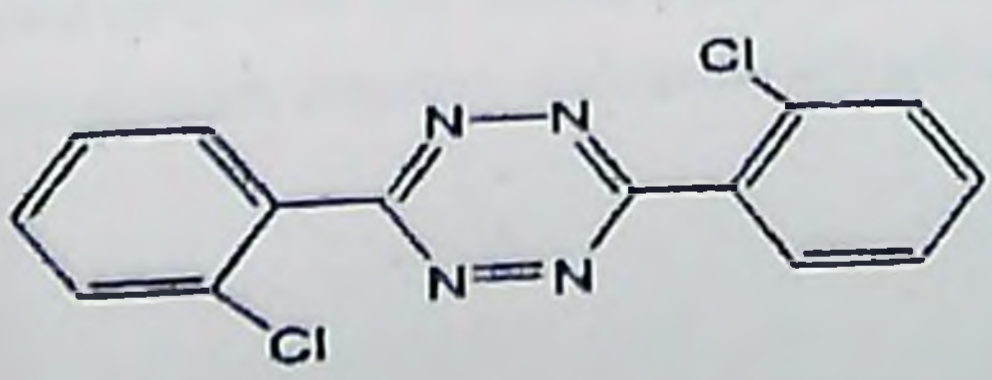


3 (RH-5992, tebufenozide)

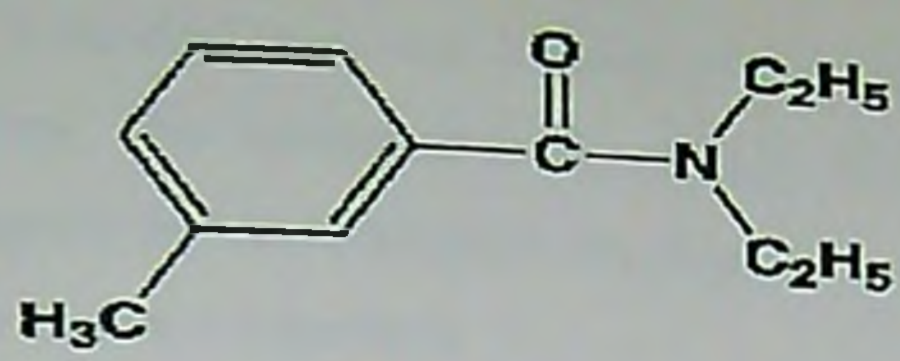
MISCELLANEOUS COMPOUNDS

New molecular structures are being synthesized routinely by basic manufacturers in search of new insecticides with new modes of action. Clofentezine (Apollo®, Acaristop®), belongs to the unique group, the tetrazines, used as an acaricide/ovicide for deciduous fruits, citrus, cotton, cucurbits, vines and ornamentals. It inhibits mite growth, but the mode of action is not known.

CLOFENTEZINE (Apollo®, Acaristop®)



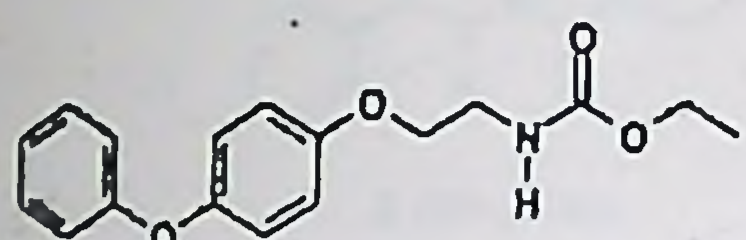
3,6-bis(2-chlorophenyl)-1,2,4,5-tetrazine



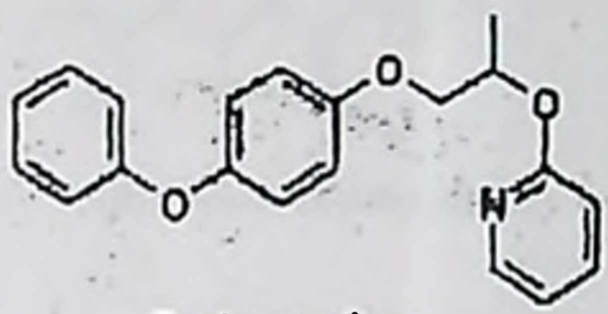
DEET-*N,N*-diethyl-*m*-toluamide

INSECT GROWTH REGULATORS

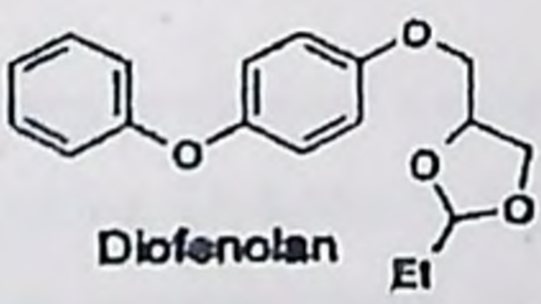
The most active ones such as methoprene and hydroprene, however lack the epoxide function present in JH. More recently, several highly active compound that have less apparent similarity to juvenile hormones have been synthesized (Tarlochan *et al.*, 1998). They are fenoxycarb (Insegar®, Logic®, Torus®, Pictyl® and Varikill®). Pyriproxyfen(Knack®, Sumilarv® and Admiral®) and Diofenolan (CGA 59205®, Aware®)



Fenoxycarb



Pyriproxyfen



Diofenolan

The first bisacylhydrazine compound was discovered by Rohm and Hass Company Scientist in 1983. RH 5849 – It has modest activity against lepidopterans, Coleopteran and dipteran pests.

RH 5992 (Tebufenozide) currently marketed as Mimic®, Confirm® and Romadam® . It is significantly more toxic than RH 5849. RH 0345 (Halofenozide) It has soil systemic activity against scarabid beetle larvae, cut worms and Web worms. This compound has been introduced for use on turfgrass and Ornamentals in USA. Trade name MACH 2® (Joint venture of Rohm and Hass company & American Cyanamid company)

Enzone®, sodium tetrathiocarbonate, is used only on grapes and citrus applied as a water application and irrigated into the soil. It breaks down in the soil to form carbon disulfide, which acts rapidly, decomposes quickly, and is effective against nematodes, soil insects, and soil borne diseases.

Clandosan® is a naturally occurring product derived from crab and shrimp shells and used as a nematicide. It is a dried, powdered, chitin protein isolated from crustacean exoskeletons and blended with urea. It stimulates growth of beneficial soil microorganisms that control nematodes, but does not have a direct adverse effect on nematodes as such..

CONCLUSION

In order to catch up with the rest of the world and become self reliant in the field of pesticides, a vigorous programme of basic research on new products is required. Even though some efforts are being made by various research agencies like CSIR, universities etc., there is no concerted effort as of now for undertaking basic research of new pesticide development. In spite of a relatively large research infrastructure and highly qualified and experienced technical manpower available, it is rather distressing to note that India has so far not contributed any single molecule for commercial exploitation as a pesticide.

Although it is inevitable that insects will ultimately develop resistance to any new insecticide particularly when it is misused, resistance to the newer molecules ought to be delayed by implementation of resistance management programmes. These compounds should be particularly suited for use in resistance and IPM Programme because of their novel mode of action and high level of safety to predators and parasitoids. Our defence against pest complex in tomorrow's world will be compounds that are economically attractive, ecologically safe and user friendly.

DISCUSSIONS

1.Who is responsible for monitoring the insecticide rules in Kerala?

Agricultural officers, Department of Agriculture is responsible for monitoring the insecticide rule in kerala.

2.Whether Endosulfan is banned in our country?

No, It is not yet banned in our country. It is under investigation. But in Kerala it is banned.

3.What is the period for temporary banning of insecticides?

Temporary banning of insecticide is valid for 30 days.

4.Whether Fiproles are available in India?

Yes It is available in the trade name of Regent. It is used for seed treatment and formulated as baits for cockroaches, ants and termites

5.Whether Fenpyroximate is effective against mite under Kerala condition?

Yes Fenpyroximate 5EC provide significant superior control of eriophyid mite *Aceria guerreronis* @ 10 ml/ palm in the case of root feeding and 0.75 –1.0 ml / liter of water as direct spraying and is much less toxic to predatory phytoseid mite *Amblyseius sp.*

6. What is the mode of action of Benzoyl ureas?

The benzoylureas act on the larval stages of most insects by inhibiting or blocking the synthesis of chitin, a vital and almost indestructible part of the insect exoskeleton. Typical effects on developing larvae are the rupture of malformed cuticle or death by starvation. Adult female boll weevils exposed to diflubenzuron lay eggs that do not hatch. And, mosquito larvae control can be achieved with as little as 1.0 gram of diflubenzuron per acre of surface water.

8.What is the mechanism of action of insect growth regulators?

Insect growth regulators interfere with growth and development of an insect. JH analogs and ecdysteroids interfere with moulting and metamorphism of insects results in abnormal larvae, pupae and adults . It affects fecundity and reproduction also.

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IATROGENIC PLANT DISEASES

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SEMINAR REPORT

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ABSTRACT

Iatrogenic is a term used in human medicine for " diseases causes by treatment especially for some other ailment". Horsfall (1972) introduced this term in plant pathology. It refers to those diseases that results from the use of plant protection chemicals or diseases that are increased in severity following the use of these chemicals.

Iatrogenic plant diseases are grouped in to three depending upon the action of agrochemicals on the host plant, the pathogen and the eco system in which the host and pathogen co-exist.

Metalaxyl formulations showed iatrogenic effect on the Alternaria leaf spot in rapeseed and mustard, when used for the control of downy mildew and white rust (Sawant & Kolte, 1985). During the evaluation of different fungicides against anthracose of grapes, a marked increase in the intensity of bacterial leaf spot was observed on plants treated with Bavistin @ 0.1 % (Mohan *et al*, 2001)

CCC, usually used to induce dwarfing in wheat increased head infection by pathogens and eye spot diseases (Bockmann, 1968). Horsfall & Dimond, (1957), reported the iatrogenic effect of 2,4-D in target spot of tomato, induced by *Alternaria solani*.

Hence for better crop production it is necessary to assess the whole protection program. In the case of iatrogenic plant diseases integrated control is the only possible way to reduce their deleterious effect.

IATROGENIC PLANT DISEASES

INTRODUCTION

Crop production is essentially an exercise in the management of artificial plant ecosystems. The objective is to achieve maximum yield of valuable produce per unit area of land. Thus optimum conditions for growth and development of crop is necessary, which include freedom from competing plant species, pests and diseases. Crop protection i.e, control of weeds, pests and diseases using agrochemicals, herbicides, growth regulators, insecticides, fungicides etc. is an important element in crop production system. Although these chemicals are necessity, if used with out careful selection, they can effect on non target organisms or physiological process. This include the induction of new diseases or the exacerbation of diseases already present.

Diseases which result from or are increased in severity by the use of specific crop protection chemical are referred as iatrogenic plant diseases.

'Iatrogenic' is a term used in human medicine for 'a disease caused by treatment, especially for some other ailment'. For example, radioactive Iodine used in the treatment of hyperthyroidism results in hypothyroidism and this condition is regarded as iatrogenic. Again streptomycin used in the treatment of Tuberculosis can cause vestibular disturbances or ototoxicity, which is also iatrogenic. Calcium channel blockers such as niphidipin and amylodipin used in the treatment of hypertension causes iatrogenic oedema.

In plant pathology, usually diseases induced or increased due to agrochemicals are considered as iatrogenic. Horsfall (1972) indicated that this term is equally applicable in plant pathology. Main symptoms of iatrogenic plant diseases are:

- herbicide applications can stimulate plant roots to leak metabolites making them more susceptible to root pathogens
- fungicide applications which eliminate antagonists and hyperparasites of plant disease fungi

Latrogenic diseases are becoming increasingly important as our usage of crop protection chemicals grows. Ofcourse, there is increased need of applying crop protection chemicals in the crop management system. So there is also a need of testing the crop protection chemical, to be aware of their side effects, including their potential to cause iatrogenic diseases. Here, the topic is gaining more and more importance.

CLASSIFICATION

Iatrogenic diseases can be divided into three broader groups according to which the disease is induced or exacerbated through action of agrochemicals on

a) The host plant

b) The pathogen

c) The ecosystem in which host and pathogen coexist

(Griffiths and Berrie, 1978)

A) THE EFFECT OF HOST PLANTS

There are no instances where the toxic principles in insecticides or fungicides have been responsible for changes in the structure or physiology of host plants. Emulsifiable insecticide formulations were shown by Natti and Hervey (1956) to increase *Perenospora parasitica* on broccoli. Here emulsifying agents dissolve the waxy bloom on the leaves rendering them more susceptible to infection.

By contrast to this, the use of herbicides and growth regulators have commonly been associated with increased disease. Various mechanisms involved are grouped into three main categories:

1. Changes in host composition and structure
2. Leakage of metabolites to the host surface
3. Changes in natural defence mechanisms

(Katan and Eshel, 1973)

1) **Changes in host composition and structure**

Horsfall and Dimond (1957) showed that 2,4-D and maleic hydrazide can induce changes in sugar concentration with in host tissues; here MH increases and 2,4-D decreases the sugar content of leaves. Thus attained low or high sugar condition is preferred by certain fungi. Target spot of tomatoes induced by *Alternaria solani*, a low sugar fungi, is increased by 2,4-D and decreased by MH. By contrast rusts, high sugar fungi, are decreased by 2,4-D and increased by MH.

Simazine, has another type of effect, it is known to increase the nitrogen content of plants. Here increased nitrogen in the host tissue is positively correlated with increased susceptibility of diseases. So its application increases the severity of many diseases, for example chocolate spot of field beans, maize smut, sugarcane mosaic and powdery mildew in cereals (Ries, 1976).

Botrytis rot of grapes, tomatoes and strawberries is increased by application of ethylene bis (dithiocarbamate) fungicides. The reason is that they can disturb mineral nutrition in the host (Stall, 1963).

Chlormequat chloride (ccc) induces dwarfing in wheat and thus increases the effects of eyespot induced by *Pseudocercosporella herpotrichoides*. It's use can increase head infection by both *Septoria nodorum* and *Fusarium culmorum*. By a series of experiments carried out by Bockmann (1968) found that yield were regularly reduced. He suggested that the yield reduction is related to the dwarf habit induced by ccc or ccc is the major determinant in yield reduction.

2. Leakage of metabolites to the host surface

Soil applied herbicides are widely used in the cultivation of many crops. They are often present in the soil at their highest concentration, when crops are at the seedling stage. Their use exacerbates seedling diseases caused by fungal pathogens, usually species of *Rhizoctonia*, *Fusarium*, *Pythium* and *Phytophthora* (Griffiths, 1981).

Juvenile tissues are more vulnerable than mature ones because natural defence mechanisms are often poorly developed at this time. Here an additional factor contributes to greater susceptibility i.e., by leakage of host metabolites to the surfaces of roots and hypocotyls. This is a natural phenomenon which leads to stimulation of pathogen growth in immediate vicinity of the seedling (Campbell and Altman, 1976).

Many herbicide treatments which increases disease in seedlings have been shown to increase leakage of metabolites. Seedling blight of sugarbeet caused by *Rhizoctonia solani* is increased by several herbicides, for example cycloate. Here leakage of metabolites is considered as an important contributing factor (Altman, 1972). Picloram increases root rots of wheat and corn seedlings and the increase in disease is positively correlated with increased leakage of sugars (Lai and Semenuik, 1970). They concluded that increased carbohydrate exudation from cereal seedling may account for increased root damage from soil borne root pathogens in picloram treated soil.

3. Changes in natural defence mechanisms

Plant growth regulators and herbicides can modify natural defence mechanisms in plants.

Cunningham (1953) reported increased *Fusarium* infection of potato tubers followed by treatment with methyl esters of NAA. This treatment actually retards wound healing and thus prolongs the period of susceptibility. In peas, the use of trichloroacetic acid (TCA), as preemergence herbicide could reduce the amount of cuticular wax on leaves, leaving them particularly susceptible to damage by dinoseb and subsequently to infection by *Ascochyta pinodella* (Rademacher, 1959). Field observations by Romig and Sasser (1972) indicated increased disease due to *Rhizoctonia solani* on snapbean when trifluralin or dinoseb was applied to soil. They found that the chemical could reduce cellulose content, pectin content and resistance to mechanical penetration of hypocotyls. Also amount of phytoalexins in bean plants was reduced in direct proportion to the concentration of herbicide applied.

B) EFFECT ON PATHOGENS

Agrochemicals may stimulate growth of pathogen by selectively inhibiting competitive or antagonistic organisms. Also growth or reproduction of the pathogen may be enhanced.

Partyka and Mai (1968) reported an increase in Sclerotinia rot of lettuce following treatment of soil with the fumigant biocides like DD, nemagone, telone. The reason is that these compounds stimulated apothecial development, thus increasing the inoculum available for infection.

Root rot of navybeans, induced by *Fusarium solani* increased by a number of herbicides. Wyse *et al.* (1976) found complex interactions between host, pathogen, environment and herbicide. With one herbicide, atrazine, stimulation of *Fusarium solani* was increased four fold. Also other herbicides such as alachlor, chloramben, EPTC etc. increased the same disease. Here spore germination, growth of germtubes and subsequent formation of chlamydospores were enhanced in soil with herbicide application.

Stimulation of spore germination by low concentration of fungicides has been reported by Nutman and Roberts (1962). Uredospores of *Hemileia vastatrix*, the causal agent of coffee leafrust, germinated more readily in the presence of low concentrations of copper (1 ppm). Here residual fungicide on coffee leaves increased the infection.

Sawant and Kolte (1985) reported iatrogenic effect of metalaxyl on Alternaria leaf spot of rape seed and mustard. Metalaxyl formulations Apron 35 SD and Ridomil 25 WP, when used in field trials as seed treatment and spray respectively, for the control of downy mildew and white rust of rapeseed and mustard, increased severity of Alternaria leaf blight. Both number and size of spot increased in fungicide treated plants. Increased sporulation was found with increased concentration of fungicide. In *in vitro* experiments using the fungicide also, growth and sporulation of *Alternaria brassicicola* and *Alternaria bassicae* was increased. Fungicides could not alter the viability of spores and also germination of spores.

During evaluation of different fungicides against anthracnose of grapes in 1992 and 1993, a marked increase in the intensity of bacterial leaf spot by *Xanthomonas campestris* pr. *viticola* was observed on plants treated with Bavistin @ 0.1% (Chander Mohan *et al.*, 2001).

The intensity of bacterial leaf spot showed a marked increase on the plants treated with Bavistin @ 0.1%. It was 52.8% in the treatment with Bavistin compared to 41.5% in the untreated check. In contrast, it was drastically reduced to 8.5% by sprays of BM. In treatments where Bavistin sprays were alternated with BM, an appreciable decrease in bacterial leafspot intensity was recorded.

EFFECT OF BAVISTIN AND OTHER FUNGICIDES ON THE INTENSITY OF BACTERIAL LEAF SPOT IN A CHEMICAL CONTROL TRIAL ON ANTHRACNOSE OF GRAPES

Fungicide	Dose (%)	Disease Intensity (%)	
		Anthracnose	Bacterial leaf spot
Bordeaux mixture	0.8	5.3	8.5
Dithane M-45	0.3	9.2	24.0
Bavistin	0.1	3.5	52.8
Bavistin/BM	0.1/0.8	4.7	18.7
Phosphoric acid	0.15	21.2	38.8
Captaf	0.25	10.5	13.2
Control	-	24.0	41.5

(Chander Mohan, Thind and Soni, 2001)

In artificial inoculation condition also, Bavistin (0.1%) provided similar effects. Also Bavistin at any of the concentrations tested, promote the growth of *Xanthomonas campestris* pv. *ificola* and not inhibit its growth. So these results show that carbendazim has an Iatrogenic effect against the bacterial leaf spot of grapes. In this case, control of anthracnose organism by carbendazim may be an important factor, but lead to increased severity of bacterial leaf spot later.

Iatrogenic effect of copper oxychloride (Blitox) on Alternaria blight of crucifers by *Alternaria brassicae* was reported by Thind and Jhooty (1985). The effect may be the result of utilization of low concentrations of copper left as residue on host, used by *Alternaria brassicae* for its germination and further development.

C) EFFECTS ON THE ECOSYSTEM

The survival of many pathogens outside the host is strongly influenced by competitive or antagonistic organisms. Those treatments which selectively inhibit such organisms will tend to increase the abundance of the pathogen in the environment, thus increases disease intensity and severity.

Vanden Berg and Bollen (1971) showed that Aster wilt induced by *Phytophthora cryptogea* increased after treatment of soil with fungicide benomyl. Here after application of benomyl, there will be an increase in benomyl tolerant organisms which include *P. cryptogea*.

A more complex situation was the action of chloropicrin and quintozene on *Phytophthora cinnamomi* causing root rot of pineapple (Baker and Cook, 1974). Chloropicrin controlled the disease for three years, by contrast quintozene, which is equally toxic to *P. cinnamomi*, increased disease. The reason is that antagonists of *P. cinnamomi* mainly *Trichoderma viride* were stimulated by chloropicrin whereas they were depressed by quintozene. Since *T. viride* is antagonistic to many root pathogen, quintozene application could exacerbate other root diseases. Therefore Pythium root rot of pine seedlings and root rots of various hosts caused by *Fusarium* and *Helminthosporium* are increased by quintozene application.

Biological control is not restricted to soil pathogen. Many diseases effecting aerial parts of plants are inhibited by competitors, antagonists or hyperparasites where such biological control is operative, it is a reasonable expectation that fungicides will create, rather than solve diseases.

First example is Botrytis rot of cyclamen corms where benomyl was used to control the disease (Bollen, 1981). Initially it was highly effective, but resulted in a situation where the disease was much more severe on treated than on untreated plants. Here mutant forms of *Botrytis cinerea*, highly resistant to benomyl soon become dominant in the population. On these two strains of *Pencillium brevicompactum* were found antagonistic to *B. cinerea*. Initially infection of corms by *B. cinerea* was checked by fungicide. The use of benomyl again has controlled both fungi i.e., antagonistic and pathogenic fungi. But the emergence of benomyl tolerant strains of *B. cinerea* in the absence of naturally occurring antagonist *Penicillium brevicompactum* resulted in more infection.

Second one is dieback of Apricot caused by *Eutypa armeniacae*. It is the only major disease of apricot trees. This disease emerged as important due to the introduction of routine sprays with copper fungicides for control of shot hole disease caused by *Clasterosporium carpophilum*. Here cultures from pruned shoots which are not infected with *E. armeniacae*, frequently yield the saprophytic fungus, *Fusarium lateritium* and pre inoculation of cut shoots with this species effectively control infection by this pathogen. Equally good control achieved by spraying benomyl or thiabendazole to cut shoots. Here a potential for combining both biological and chemical control methods exists because *F. lateritium* spores are much less sensitive to benomyl and thiabendazole (Carter and Price, 1975).

Another important example is the case of leaf rust and berry disease of coffee in major coffee growing areas. *Coffea arabica* has been effected by two major diseases, leaf rust by *Hemileia vastatrix* and coffee berry disease by *Colletotrichum coffeanum*. Effective control of these diseases can be achieved only by frequent fungicide application. Ironically, in the absence of fungicides, coffee berry disease was of negligible importance and leafrust less damaging. So both diseases can be regarded as iatrogenic. But coffee growers especially in East

Africa, always used fungicides. Because a single spray of BM applied at the beginning of the 'long rains' (March-May) was found to increase the yield of berries in the following year. It has been showed that mean annual increase with one spray in March was over 100%. Because of this, 'tonic' spraying has become a standard practice. The reason behind this is that, because of these spraying there was a retardation of natural leaf fall in the dry season. So trees will be well foliated throughout the year. Again yield is linked to leaf retention because the number of flowers indicated is positively correlated with the number of leaves. Vandervossen and Browning (1978) worked on this; has concluded that the leaf fall is not natural, but caused by certain fungi and these fungi can be controlled by fungicidal application. Control of these fungi inturn, increase the disease causing fungi, in the case of rust.

In the case of *C. coffeanum* (berry disease), pathogen is abundant in the bark, the immediate spraying was to reduce sporulation of the organism. But this is short lived and 9-12 months after spraying, sporulation capacity of organisms is much increased. Consequently, the net result of spraying is the creation of a potentially more serious problem. Reasons for increase of *C. coffeanum* in the bark is considered as selective inhibition of antagonists and competitors of the pathogen (Furtado, 1969).

CONCLUSION

The existence of iatrogenic diseases cannot be refuted. Horsfall (1979) commented that he was impressed by the number of iatrogenic diseases that exist but those so far documented do not reflect the actual problem. In the past, with the emergence of each new disease, attempts were made to devise control measures which fit into existing agronomic and economic patterns. Now very high yields of wheat and barley were attainable by virtue of efficient weed control, large fertilizer applications and the use of suitable cultivars. But crops grown in this way are more susceptible to diseases and to lodging. To combat this, straw shorteners are required, but these only increase disease proneness further. But frequent chemical applications are necessary as "economic risk insurances".

As long as crop production practices remain profitable, there is no incentive to change them. Because to change, it is necessary to assess or reassess the whole protection program for a crop. Here development of an integrated control is the only possible way as in the case of dieback of apricot, which is an outstanding example. So the new control developed should not only be effective, but also less costly and less damaging environmentally. So demonstration and analysis of iatrogenic diseases can produce a clear understanding of disease problems and aid in the definition of overall strategies for control.

DISCUSSION

1. If the use of a chemical increases a disease ,whether we can ban it?

   If the use of a chemical increases the disease, we will conduct experiments to test this. After conducting experiments in field and then in lab, if we find it is causing disease, we can surely ban. But the banning should be for that particular crop only.

2. Whether there is any treatment to reduce the suppressive effect on particular antagonists?

   Only certain chemicals has suppressive effect on particular antagonists. So we can change chemical in such a way that the new chemical is effective against the disease and also tolerant to antagonists.

3. Whether iatrogenic disease symptoms are same as original disease synmptoms? Is they aggravate the disease in the case of coffee?

   Same symptoms are seen in the case of iatrogenic disease. In coffee, iatrogenic condition aggravate the disease.

4. What is the difference in the case of symptoms appear during physiological disorder and iatrogenic disease?

   Iatrogenic disease is seen only after application of a plant protection chemical. If the disease is go on increasing with the use of chemicals, we can make sure that it is iatrogenic. Again symptoms of physiological disorders are different such as fasciation, malformations etc., and iatrogenic symptoms are same as that caused by particular disease.

5. Is integrated control is practical or have been done in any iatrogenic disease, then how?

   It is only done so far in the case of dieback of Apricot, that is why it is considered as a classical example. By combining both chemical and biological methods, we can effectively control such diseases. But the chemicals we are using must be tolerant to bioagents/antagonists.

6. What is the difference in diseases occurred in a susceptible host and iatrogenic chemicals?

   In iatrogenic diseases use of chemical can induce/increase diseases but in the case os susceptible host, usually no interference of chemical is there. Actually effect of chemicals sometimes makes host susceptible to disease.

7. What can be the control for iatrogenic disease?

   Only integrated control is possible here that is by combining chemical and biological methods.

   Awareness of disease due to use of chemical and then integrated control measures is the correct way of controlling them. But chemicals should be tolerant to antagonist.

8. How farmers can identify iatrogenic diseases in field?

   Iatrogenic disease can seen only after application of a particular chemical. If the disease increases in the presence of chemical or less damaging in the absence of chemical, that will be iatrogenic.

9. How the chemicals suppress antagonists?

   Actually they can increase the growth and development of pathogen, thereby suppressing antagonists or directly suppress the growth of biocontrol agents.

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USE OF RHIZOBACTERIA IN PLANT DISEASE MANAGEMENT

799

By

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SEMINAR REPORT

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ABSTRACT

Recently disease management using biocontrol methods is gaining momentum due to its non-hazardous and long-term effects. Among the various approaches of biological control, the use of rhizobacteria as a biocontrol agent is emerging as a popular trend, due to its additional benefits of promoting growth and yield (Kloepper *et.al.*, 1989)

The first documented work was done in 1972 using *Agrobacterium radiobacter* strain K-84 to suppress the pathogen *Agrobacterium tumefaciens* causing crown gall disease in stone fruits. (Kerr, 1972). Later several other rhizosphere bacteria belonging to *Actinoplanes*, *Alcaligenes*, *Arthrobacter*, *Azobacter*. *Bacillus*, *Bdellovibrio*, *Enterobacter*, *Flavobacterium*, *Nocardia*, *Pseudomonas*, *Rhizobium*, and *Serratia* were also used as biocontrol agents (Pal and Jalali, 1998). Among these *Bacillus* and *Pseudomonas* were commercially well exploited and they were found to suppress wide range of pathogens.

Normally these bacteria suppress the pathogens by various mechanisms such as competition for nutrients, Siderophore production, antibiosis, parasitism and induced systemic resistance.

INTRODUCTION

According to the recent WHO estimates, approximately 7,50,000 people are taken ill every year with pesticide poisoning and up to 14,000 of these are died in agony. Pesticides are necessary at present, but are not a long-term solution to crop health. Besides their non-target effects and hazardous nature, petroleum based pesticides will become more expensive, and some are now loosing their effectiveness because of development of resistant strain. Breeding for resistance, which continues to be the most practical and feasible method to control plant diseases, is not able to keep pace with the development of more virulent pathogens.

There is considerable pressure from public and environmental scientist to decrease emphasis on chemical control. So as an alternative and as a part of Integrated Disease Management now biological control is gaining momentum. Biological methods mainly consist of using a microorganism to control harmful microorganism causing plant diseases without disturbing the ecological balance. Antagonistic fungi, bacteria and mild strains of virus are employed as bio-control agents. Recently Rhizobacteria are effectively utilized in Plant Disease Management.

RHIZOSPHERE: It is a zone of increased microbial activity in the immediate vicinity of plant roots. The rhizosphere is very dynamic environment and it harbors variety of microorganisms. The rhizosphere bacteria that can colonize the roots have been termed as rhizobacteria (Kloepper and Schroth, 1978).

Classification: These rhizobacteria are classified as harmful and beneficial. Rhizobacteria that are harmful for growth of plants are called deleterious rhizobacteria (Schippers and Baker, 1987).

Beneficial Rhizobacteria include two main groups of bacteria.

I. One which establishes symbiotic relationship

E.g. Rhizobia and Brady rhizobia

These bacteria have been used extensively world wide as legume inoculants for increasing crop yield.

2. Plant Growth Promoting Rhizobacteria (PGPR)

These bacteria generally

- Produces plant growth promoting substances
- Increases the availability and uptake of minor nutrients
- Suppress soil borne pathogens and other deleterious microorganism
- Induces resistance in the plant

Rhizosphere bacteria as bio-control agents

The rhizosphere bacteria are ideal for use as bio-control agents as they can provide the front line defense for plant roots against the attack by various plant pathogens.

Sanford, Millard and Taylor did the initial work of using rhizobacteia as biocotnrol agents during 1920. The demonstrated that Potato scab caused by *Streptomyces scabies* was controlled by using *Bacillus* sp and *Pseudomonas fluorescens*. However the well-documented work was done in 1972 by using *Agrobacterium radiobacter* strain K-84 that will suppress the pathogen *Agrobacterium tumefaciens* causing crown gall disease (Kerr, 1972). Major rhizobacteria used as biocontrol agent are shown below.

List of Rhizobacteria used as biocontrol agents

Bio-control agents	**Suppressed pathogen**
Actinoplane sp	*Phytophthora*
Agrobacterium radiobacter	*Agrobacterium tumefaciens*
Alcaligenes	*Agrobacterium tumefaciens*
Alcaligenes	*Fusarium, Pythium*
Arthrobacter sp	*Pythium debaryanum*
Azotobacter chroococcum	*Rhizoctonia solani*
Bdellovibrio bacteriovorus	*Pseudomonas syringae pv glycinae*
Enterobacter cloacae	*Pythium sp, Rhizoctonia solani*

Erwinia herbicola	*Erwinia amylovora*
Flavobacterium sp	*Rhizoctonia solani*
Hafnia sp	*Fusarium oxysporum* f. sp. *dainthi*
Micromonospora sp	*Phytophthora sp*
Serratia marcescens	*Cucumber mosaic virus* *Fusarium oxysporum* f.sp. *cucumerinum*
Pseudomonas	*Rhizoctonia solani* *Pythium sp* *Erwinia amylovora* *Fusarium oxysporum* *Gaumannomyces graminis tritici*
Bacillus sp	*Rhizoctonia solani* *Pythium sp*

Source: Pal and Jalali, 1998

Among these, three bacterial species *Pseudomonas*, *Bacillus* and *Agrobacterium* were successfully tested and commercially exploited in the field.

Agrobacterium radiobacter strain K- 84

It was the first bacterium, which was used successfully as bio-control agent against crown gall disease caused by *Agrobacterium tumefaciens*. The bacterial strains with bio-control potential were isolated from symptom less plants growing in diseased field and identified as *Agrobacterium radiobacter*. Antagonistic action of *Agrobacterium radiobacter* was due to production of bacteriocin called Agrocin – 84. These bacteriocin encoded by a plasmid p Ag 84 are responsible for antagonism towards the pathogen *Agrobacterium tumefaciens* that appears to compete for the same niche on the rhizoplane.

Seed and root inoculation with *Agrobacterium radiobacter* strain 84 controlled 98.6% of crown gall of stone fruit. (Kerr, 1972).

Bacillus sp.

Several sp of the genus Bacillus have been found predominantly in the rhizosphere of various crops. In general, the genus represents the most utilized biocontrol agents because of its inherent superiority in biological control. The resistant endospores that *Bacillus* sp. provide tolerance to heat and cold as well as to pH extremes, pesticides, fertilizers and storage. In addition the application of some *bacillus sp.* have shown increased grain yield and plant biomass accumulation. The following are the examples of various bacillus sp. used as biocontrol agents.

***Bacillus* sp. as biocontrol agents**

Bacillus species	Suppressed pathogen
Bacillus cereus	*Alternaria solani* *Pythium aphanidermatum*
Bacillus polymyxa	*Gaumannomyces graminis var tritici* *Ralstonia solanacearum*
Bacillus pumilus	*Gaumannomyces graminis var tritici*
Bacillus subtilis	*Fusarium sp* *Rhizoctonia solani* *Sclerotium rolfsii*
Bacillus uniflagellates	Tobacco mosaic virus

Bacillus subtilis strain A-13 isolated from the lysed mycelium of *Sclerotium rolfsii* (Broadbent *et al.*, 1971). It was found inhibitory to several plant pathogens and improved the growth of many plant species. This strain when used as seed treatment increased the yield of carrot by 48%, oats by 33% and peanuts up to 37% (Merriman *et al.*, 1974).

Jubina and Girija (1998) conducted an experiment on efficacy of bacterial antagonists in the management of foot rot of black pepper caused by *Phytophthora capsici*. They selected 8 isolates of *Bacillus sp* from different pepper growing areas of kerala for the bio-control of foot rot. The three bacterial isolates B_5, B_7 and B_{13} were effective in checking the disease and reducing mortality to 43% and 57% compared to 100% in control after 17 days of inoculation. They provided prolonged protection to the plants even after 90 days of application. Isolate B_{13} was identified as most promising in reducing mortality. The results of the experiment are shown in table 1.

Table 1. Efficacy of Rhizosphere Bacteria (Isolated From Different Pepper Growing Areas) in Management of Foot Rot of Black Pepper (Jubina and Girija, 1998)

Bacterial isolates	% Mortality (Days after inoculation)				
	15	17	30	60	90
B3	42.86	71.43	71.43	85.71	85.71
B4	28.57	42.86	85.71	85.71	100
B5	14.29	42.86	71.43	71.43	71.43
B6	28.57	85.71	85.71	85.71	100
B7	42.86	42.86	71.43	71.43	71.43
B12	57.14	100	100	100	100
B13	42.86	57.14	57.14	57.14	57.14
B14	57.14	57.14	85.71	100	100
Control	71.43	100	100	100	100

Studies on growth promotion of different bacterial isolates showed that B_7 and B_4 enhanced the growth of pepper plants. Isolate B_7 had the dual function of disease suppression as well as growth promotion (Table 2).

Table 2. Effect of bacillus on shoot length and leaf number of black pepper (Jubina and Girija, 1998)

Bacterial isolate	Shoot length (cm)			Leaf Number		
	Days after inoculation			Days after inoculation		
	30	60	90	30	60	90
B3	23.63	38.60	54.30	6.67	8.67	10.33
B4	32.27	62.50	94.57	7.67	10.67	13.67
B5	35.93	58.97	74.17	7.00	9.33	10.67
B6	29.63	48.17	56.90	6.67	8.67	10.33
B7	29.83	64.77	82.73	8.00	12.00	15.00
B12	31.47	57.93	80.70	7.33	10.67	12.67
B13	25.40	43.23	58.00	5.67	7.33	7.67
B14	27.4	55.57	84.53	5.67	8.67	9.67
Control	16.73	24.53	41.50	2.67	5.00	6.67

Pseudomonas as biocontrol agents

The worldwide interest in the *Pseudomonas* sp as biocontrol agents was sparked by the studies conducted at the University of California, USA during late 1970's. Presently they seem to be one of the appealing candidates for the biological control of plant diseases. Several species of Pseudomonas were used as biocontrol agents.

Pseudomonas species used as biocontrol agents

Biocontrol agents	**Suppressed Pathogen**
Pseudomonas aureofaciens	*Gaumannomyces gramins* var *tritici*
Pseudomonas corrugata	*Monilinia sp*
Pseudomonas fluorescens	*Fusarium oxysporum* *Pythium ultimum* *Phytophthora sp* *Sclerotium rolfsii* *Rhizoctonia solani* *Xanthomonas campestris* pv *oryzae*
Pseudomonas putida	*Erwinia carotovora*
Pseudomonas aeruginosa	*Rhizoctonia solani*

Among the Pseudomonas species, Fluorescent Pseudomonads make up a dominant population in rhizosphere and possess several properties that have made them as biocontrol agents. These include:

- Efficient colonization of roots, tubers and hypocotyls
- Ability to utilize large number of organic substrates commonly found in root and seed exudates
- Easily cultured in the laboratory condition
- Production of variety of secondary metabolites which are toxic to other bacterial and fungal plant pathogens
- Compatibility with other commonly used pesticides and other biocontrol agents
- Possess plant growth promoting activity

A study conducted by Rangeswaran and Prasad (2000) on suppression of Sclerotium rot of sunflower by different antagonists showed that the number of disease free plants (63%) was highest in *Pseudomonas fluorescens* and *Pseudomonas putida* seed treatment, which was almost the same as that of fungicide treated plots (Table 3).

The bacterization of peanut with *Pseudomonas fluorescens* reduced the disease severity due to *Rhizoctonia solani* with an increase in yield by 50% under field condition (Savithry and Gananamanickan, 1987).

Table 3. Suppression of *Sclerotium rolfsii* rot of sunflower by antagonists

Isolates	Plant stand %	
	Seed treatment	Soil treatment
Pseudomonas fluorescens	60	20
Pseudomonas spp	37	20
Alcaligenes odorans	27	27
Pseudomonas putida	63	30
Streptomyces sp	43	27
Bacillus sp	13	20
Control (Infected)	17	17
Fungicide treated	67	67

(Rangeswaran and Prasad, 2000)

Works conducted at KAU

According to study conducted by AICVIP at COH, Vellanikkara, 1997-99, it was found that seed as well as soil treatment of *Pseudomonas fluorescens* and *Azotobacter chroococcum* were effective against damping off diseases of solanaceous vegetables like tomato, brinjal and chilli.

A work was done by Saiffuneesa, 2002 on Management of Sheath Blight of Rice. *Rhizobacteria belonging to genera Clostridium, Sporosarcina, Pseudomonas* and *Bacillus* were isolated and found antagonistic to the pathogen

Rhizoctonia solani. Among this *Bacillus* and *Sporosarcina* were found to inhibit sclerotia formation of pathogen.

A pilot study on the efficiency of *Pseudomonas fluorescens* against sheath blight of rice was conducted during the year 1996 as a part of the AICRIP at the RARS, Pattambi. The bacterial formulation was applied as seed treatment @ 10g/kg seed followed by three foliar sprays at 30,60 and 90 days after transplanting. This was found to be effective in reducing the sheath blight incidence.

Mechanism of Disease Suppression

An understanding of the plant microbe ecology and mechanism of action of the bio-control agents is a pre-requisite for their effective use in plant disease control strategy.

Different mechanisms have been proposed to explain how rhizobacteria suppress the diseases. This include

1. Competition for nutrients
2. Parasitism and lysis
3. Siderophore production
4. Antibiotics
5. Bacteriocins
6. HCN production
7. Phytoalexins
8. Other cellular metabolites
9. Induced systemic resistance

1. Competition for nutrients and ecological niche

Competition for nutrients, supplied by root and seed exudates occurs between rhizosphere bacteria and plant pathogens. Here the fast growing rhizosphere bacteria would effectively compete with slower growing plant pathogens for these available nutrients in root exudates in the rhizosphere. Rhizobacteria establishes on roots and utilizes the nutrients available in rhizosphere and reduce the quantity available to the pathogen. Eg, The root colonizing population of *Arthrobactor spp.*

reduced the amount of available carbon and nitrogen required to stimulate the germination of oospores of the pathogenic fungi *Pythium aphanidermatum* causing damping off disease (Elad and Chet, 1987).

Ecological niche: Infection site or ecological niche has also been suggested as an important mechanism of bio-control. Usually, the points of emergence of lateral roots and cell functions of roots appears to be favoured for colonization by many kinds of rhizosphere bacteria due to abundant availability of nutrients in root exudates. The application of bacterial bio-control agents prevents the establishment of the pathogen at these sites. Eg. Bio-control of *Pythium ultimum* has been reported by the seed bacterization with antagonistic *Pseudomonas spp.* This antagonist competes for infection site with the pathogen and thus protects the sugar beet seedlings form *Pythium ultimum*

2. Parasitism and lysis

It is a mechanism by which destruction of plant pathogen occurs by the action of bio-control agents. Rhizobacteria degrade the cell wall of pathogenic fungi and certain pathogenic bacteria by producing hydrolytic enzymes like proteases, gluconases, lipases and chitinases. Rhizosphere bacteria, which control bacterial and fungal plant pathogen by parasitism and lysis are presented below.

Parasitic and lytic rhizosphere bacterial bio-control agents

Bio control agents	Suppressed pathogen
Bdellovibrio bacteriovorus	*Pseudomonas syringae pv. glycinea*
Arthrobacter spp.	*Fusarium spp. Pythium spp.*
Enterobacter cloacae	*Pythium spp.*
Serratia marcescens	*Sclerotium rolfsii*
Pseudomonas fluorescens	*Phytophthora infestans*
Bacillus spp.	*Gaumannomyces graminis var tritici*

Sherff (1973) demonstrated that an isolate of *Bdellovibrio bacteriovorus* obtained form rhizosphere of soybean prevented the symptoms of soybean blight caused by *Pseudomonas savastanoi* pv *glycinea* by parasitic action.

Bdello vibrio spp. bring about the destruction of Gram – ve bacteria by attaching to the host cell wall which they penetrate and enter the space between the cell membrane and cell wall . The rod shaped host cell is converted into spherical body. The *Bdello vibrio* cell settles in the periplasmic space and does not enter into cytoplasm of the host cell. The cytoplasm of the host cell is broken down by proteases produced by *Bdello vibrio* and is used as nutrients for its growth. The lytic enzymes, glucanase and peptidase dissolve amino sugars and other cell wall constituents of the host cell.

3. Siderophore production

In oxygenated and weakly acidic, neutral or alkaline PH of soil (PH > 6) iron (Fe^{3+}) is found as insoluble iron complexes, $Fe(OH)_3$ and it become unavailable and limiting factor for growth. To sequester the scarcely available iron, microorganisms produce low molecular weight compounds called Siderophore.

Siderophore are extra cellular low molecular weight (500-1000 Dalton) high affinity iron chelators, usually produced by several bacteria under iron stress condition to transport iron in to bacterial cell.

Siderophore mediated competition is considered as one of the important mechanisms of bio-control of plant pathogens as they create an artificial shortage of Fe for respective plant pathogens. Most of the bacteria and fungi produce Siderophore, which give competitive edge to the organisms producing stronger Siderophore having higher affinity for Fe, which will make the pathogen deprived of iron.

Kloepper and coworkers (1980) was the first to demonstrate the Siderophore-mediated competition as a mechanism of bio control especially in Pseudomonas. They showed the involvement of Siderophore in the suppression of *Fusarium oxysporum* and *Pythium spp.*

Some of the important Siderophore produced by *Pseudomonas spp.* are Pyochelin, Pyoverdin, Pseudobacteria and Ferribactin.

Production of antibiotics

Antibiotics are the substances produced by the microorganisms or plants that in very dilute concentration has the capacity to inhibit the growth of other microorganisms.

Thomas and Weller (1990) first proved that an antibiotic was involved in disease suppression. When they used Pseudomonas florescence strain 2-79 to control *Gaumannomyces graminis var. tritici* causing take all decline of wheat. The suppressive activity of this strain was attributed to the ability of this antagonist to produce an antibiotic, Phenozine –1-carboxlylic acid.

In general Fluorescent Pseudomonas are considered to be most promising among bacterial bio control agent as they produce a wide spectrum of inhibitory compounds like antibiotics. Antibiotics produced by various *Pseudomonas* spp. and the suppressed pathogen are shown below.

Antibiotic production and disease suppression

Antibiotics	Bacterial strain	Disease suppressed
Phenazine-1-carboxylic acid	*P.fluorescens* 2-79	Take all decline of wheat
2,4- Diacetyl phloroglucinol	*P.fluorescens* CHAO	Black root rot of Tobacco
DAPG	*P.fluorescens* F113	Sugar beet damping off
Oomycin A	*P.fluorescens* HV37a	Damping off of cotton
Phenyl pyrrole	*P.fluorescens* Pf 5	Damping off of cotton

Bacteriocins

The proteinaceous antagonistic substance produced by several species of bacteria that are lethal to other strain of closely related bacteria are bacteriocins.

These are adsorbed to the receptor on the surface of bacterial cell, eliminate electric charges of the inner membrane and stop biosynthesis of adenosine tri phosphate. Consequently, the biosynthesis of DNA, RNA, proteins and phospholipid is disrupted and finally the growth of bacterial cell is inhibited. Eventually it may cause cell death.

Eg. Agrocin 84 is a bacteriocin produced from *Agrobacterium radiobactor* that involved in biocontrol of *Agrobacterim tumefaciens* causing crow gall disease of stone fruits (Kerr, 1972). Agrocin 84 is a substituted adenine nucleotide. It seems to be transported in to sensitive strain of *A. tumefaciens* by one or more plasmid-encoded binding proteins located on the cell envelop. The genes for the synthesis of agrocin 84 are located on a plasmid pAg84 of *Agrobacterium radiobactor*.

Hydrogen Cyanide production (HCN)

Hydro cyanic acid is know to be produced by many rhizosphere bacteria and its role in the biological control of plant pathogens has also been demonstrated (Schipper, 1987)

P. fluorescens strain CHAO produces HCN that has implicated in the control of *Chalara elegans* in tobacco causing black root rot (Weller, 1988). HCN modifies the metabolism of tobacco plant in a way that induces some defense mechanism in the plant against the host pathogen.

Phytoalexins

Phytoalexins are low molecular weight antimicrobial compounds produced in plants in response to stress or to bacterial / fungal invasion.

Accumulations of phytoalexins in carnation plants have been reported in response to the bacterization of carnation roots with bio control agent, *Pseudomonas spp.* Strain WCS 417r. This has been found to reduce the number of disease plants infected with *Fusarium oxysporum* f. sp. *dianthi* from 50 to 20% (Van Peer *et al.* 1992).

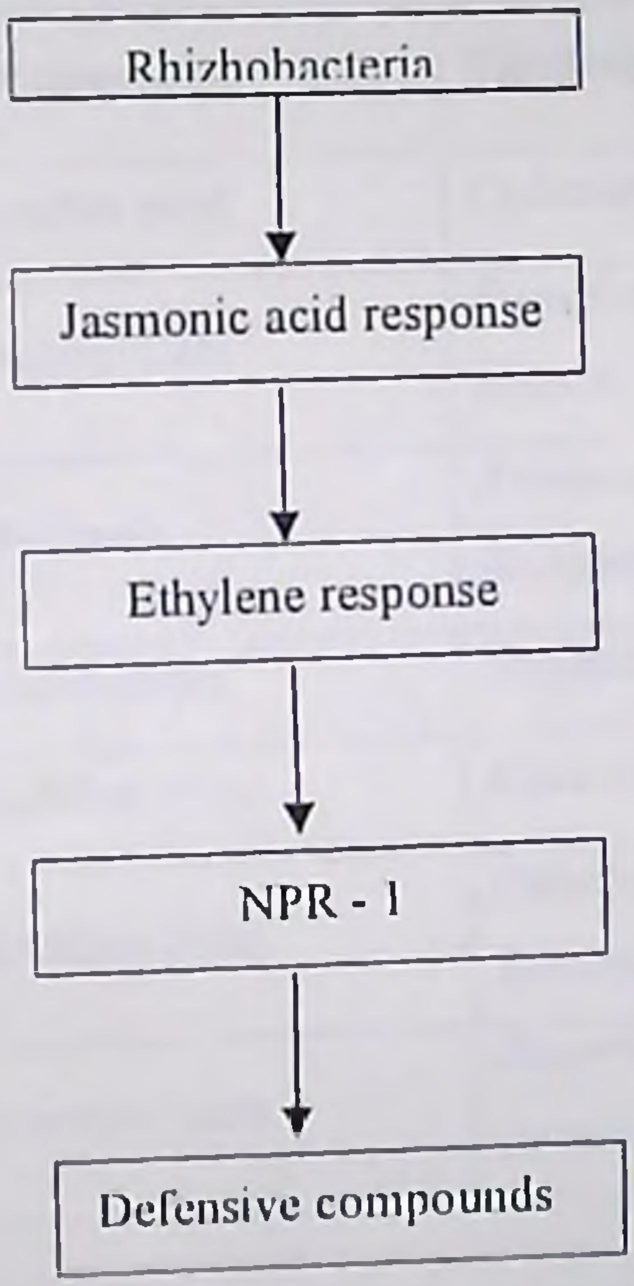
Other Cellular Metabolites

Several volatile and non-volatile compounds produced in soil atmosphere were found inhibitory to fungal spore germination. Productions of ammonia (NH_3) by *Enterobactor cloacae* have been reported to inhibit the growth of *Rhizoctonia solani* and *Pythium ultimum* (Howell *et al.*, 1988).

Induced Systemic Resistance

Most bacterial bio control agents of plant pathogens function partially or completely through antagonism. Research over the past years has demonstrated that induced systemic resistance can be an alternative mechanism of antagonism for achieving biological control of plant diseases. The rhizobacteria can suppress disease caused by foliar pathogens by triggering plant mediated resistance mechanism. This type of systemic resistance induced by non-pathogenic microorganisms has been named induced systemic resistance (ISR). (Kloepper *et al.*, 1992).

Rhizobacteria mediated induced systemic resistance



Rhizobacteria when applied induces production of a compound called jasmonic acid. High level of accumulation of jasmonate is correlated with ethylene production, which in turn trigger the synthesis of novel pathogenesis related proteins (NPR-1). NPR induces production of defensive compounds in host plants against the target pathogen

ISR once expressed activates multiple potential defensive mechanisms that include increase in activity of chitinase, β-1, 3, gluconases, peroxidases and other pathogenesis related proteins (Lawton *et al.*, 1987) accumulation of antimicrobial low molecular weight substances like phytoalexins (Kuc and Preisig, 1984) and formation of protective bio polymerase like lignin, callose and hydroxy proline rich glycoprotiens (Hammerschimidt *et al.*, 1982). Wide spectrums of pathogens are controlled with a single inducing agent. In cucumber treatment of the 1st leaf with a necrosis-forming organism protects the plant against at least 13 pathogens including fungi, bacteria and virus (Dean, and Kuc, 1985). Some of the examples for induced systemic resistance are given below.

Induced systemic resistance in plants by Rhizosphere bacteria

Inducer Bacteria	**Plant Species**	**Challenged Pathogen**
Pseudomonas spp.	Cucumber seed	*Colletotrichum orbiculare*
P.fluorescense WCS 417 r	Carnation roots	*Fusarium oxysporum* f.sp. *dianthi*
P. fluorescens WCS 417 r	Arabidopsis	*Xanthomonas campestris* pv.*campestris*
P.fluorescense CHAO	Tobacco roots	Tobacco Necrosis Virus
P. putida	Cucumber roots	Cucumber Mosaic Virus
Serratia marcescence	Cucumber roots	*Pseudomonas syringe* pv. *Lachrymans*
S. marcescence 90 – 166	Cucumber roots	*Fusarium oxysporum* f.sp. *cucumerinum*

ISR is similar phenotypically to pathogen induced Systemic Acquired Resistance (SAR) in such way that both provides enhanced resistance against challenged pathogen. SAR and ISR differ in their signaling pathways. SAR is depended on the synthesis of Salicylic Acid (SA) by the plant that acts as an Inducer signals and is associated with accumulation of pathogenesis related proteins (PR proteins), some of which have been shown to possess antifungal activity.

Pseudomonas aeruginosa 7NSK2 produces SA, which induces SAR in bean against *Botrytis cinerea* (De Meyer and Hofte, 1997).

Characteristics of an Ideal Bio control Agents

Wilson has presented certain criteria for ideal antagonists for the biological control of various plant diseases. These criteria provide a great deal of help in selecting a potential bio control agent. They are,

- Genetic stability
- High consistent efficacy
- Ability to survive under adverse environment conditions
- Effectiveness against wide range of pathogens on varieties of fruits and vegetables.
- Amenability for growth on inexpensive medium.
- Non production of secondary metabolites that might be toxic to humans
- Strain resistant to standard fungicides
- Compatibility with another chemicals and physical treatments of the commodity such as heating and waxing

Commercial Bacterial Bio control Products

Now a day's huge amount of work has carried out on the bio control of plant diseases. Even then number of commercially available bio control product is very low. This small number of products is not surprising since bio control research oriented towards product development has been pursued for a relatively short time. In India availability of commercial formulations is very less, though in other countries this is exploited very much.

Commercial Bacterial Bio control Products

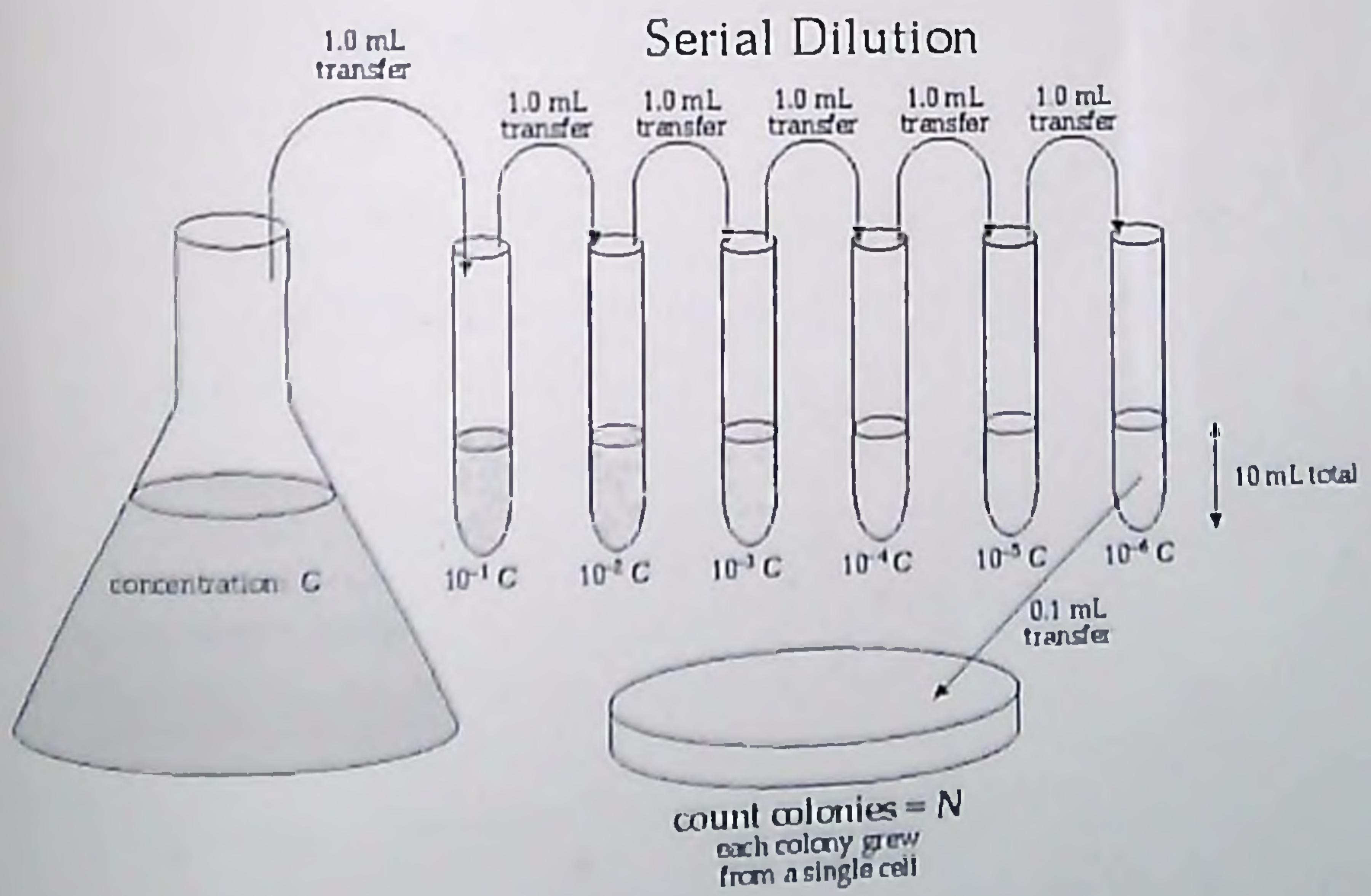
Bio control agents	Target pathogen	Product
Agrobacterium radiobacter strain K84	*A. tumefaciens*	Di egal, No gall, Galltrol
Bacillus subtilis	*Pythium ultimum* *Rhizoctonia solani*	Quantum 4000
Streptomyces griseoviridis	*Fusarium oxysporum*	Mycostop
Pseudomonas fluorescens	*Pythium ultimum* *Rhizoctonia solani*	Blight-ban Dagger-G

Isolation of Rhizosphere Bacteria from Soil

The microorganisms of rhizosphere region are isolated by serial dilution plate method.

Take 10 gm of soil along with roots from the rhizosphere region of plant. Dissolve it in 90 ml sterile water in conical flask. Gently shake the flasks using shaker for 10 minutes so as to get proper soil suspension. Serially dilute soil suspension by transferring 1 ml from conical flask to each test tube. From test tube 1 to 2, 2 to 3, 3 to 4, 4 to 5 subsequently as shown in fig to make up to 10^{-6} dilution withdraw by sterile pipette, 0.1 ml aliquot from dilutions 10^{-3} and 10^{-4} and pour on plates containing respective media. The procedure is shown in Fig. 1. Spreader is used for spreading the diluted aliquot by rotating the plate. Incubate at 30^0 C for 2-5 days. Record the visual observations after development of colonies. Kings B medium are used for isolation of Pseudomonas fluorescence.

Fig. 1. Isolation of rhizobacteria from soil



CONCLUSION

Several rhizosphere bacteria have the potential to control various root, foliage and post harvest diseases of agricultural crops caused by plant pathogenic fungi, bacteria and viruses. These bacteria are not only the appealing candidates for bio-control of plant diseases, but also many of these plays an important role in promotion of plant growth and yield.

In growing scenario of sustainable agriculture and pollution free environment any technologies that focus on minimum use of agrochemicals or environmentally safe, ecofriendly and technically feasible are always welcome. Bio-control using rhizobacteria is a step in this direction. So use of rhizobacteria in plant disease management will go a long way not only in helping the farming community by providing inexpensive and effective biocontrol agent but also in preserving the ecosystem.

DISCUSSION

1. What is the advantage of induced systemic resistance (ISR) over cross protection?

The scope of offering resistance against many species of pathogens is wide in case of ISR where as it is narrow or patho specific in cross protection. Successful cross protection system has been reported only in few crops.

2. What is the possibility of using rhizobacteria in acid soils of Kerala?

Rhizobacteria generally prefers neutral pH. So it can be used after liming or by other ameliorative measures. Alternatively native isolates of rhizobacteria obtained from acid soils can be tested fro its efficacy and suitable strains can be commercialized.

3. Is there any commercial formulation of rhizobacteria available in Kerala and what is its price?

Yes, *Pseudomonas fluorescens* formulations are available from Bio-control Lab Mannuthy and it costs about Rs. 50/ 500g.

4. Can bacterial disease be successfully controlled by using rhizobacterium.

Agrobaterium radiobacter strain K-84 was very effective in controlling crown gall disease of stone fruit caused by *Agrobacterium tumefacience.* Bacterial diseases like soft rot of potato caused by *Erwinia carotovora* and bacterial leaf blight caused by *Xanthomonas oryzae* pv *oryzae* have be successfully controlled by *Pseudomonas fluorescens.*

5. Is there any disease of post harvest disease controlled by Rhizobacteria?

Bacillus subtilis isolated from rhizosphere soil were found to be effective brown rot of peach and plum caused by *Monilinia fructicola.*

6. Can Rhizobium be utilized as bio-control agent?

Usually these rhizobium are utilized as legume inoculants for promoting plant growth and yield. Rhizobium in combination with Bavistin on seed treatment are found effective against Foot rot and Root rot of lentil caused by *Fusarium oxysporum*.

7. Is *Pseudomonas* compatible with other bio-control agent?

Pseudomonas in combination with other bio-control agent may lead to better control of plant diseases. Take all decline of wheat can be controlled by the combination of *Pseudomonas fluorescens* and *Trichoderma koningii* better than *Pseudomonas* alone.

8. What is the difference between Induced Systemic Resistance (ISR) and Systemic Acquired Resistance (SAR)?

ISR is rhizobacteria mediated induced systemic resistance. ISR and SAR differ in their signaling pathways. ISR dependent on the synthesis of jasmonic acid by the plant that act as inducer signals and is associated with the production of some defensive compounds in the host plant. SAR is pathogen induced systemic resistance. They depend on the synthesis of Salicylic acid and is associated with the accumulation of Pathogenesis related proteins (PR), some of which possess antifungal activity

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