AUGMENTATIVE BIOLOGICAL CONTROL IN GREENHOUSES: EXPERIENCES FROM CHINA

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ABSTRACT

To enhance biological control of insect pests in greenhouses, facilities and procedures for mass production of the parasitoids, *Eretmocerus* sp., *Encarsia formosa*, and *Trichogramma brassicae*, and the predator, *Aphidoletes aphidimyza* were successfully developed in Hengshui, Hebei province, China. Mass production of the aphelinid wasps was achieved by using different plant varieties and host insect species, as well as specific rearing procedures and techniques. Production of *T. brassicae* was greatly enhanced through the design of special devices and improved rearing techniques. Annual production of natural enemies in our institution reached 2 billion individuals. Biological control experiments conducted in sunlight greenhouses and plastic greenhouses allowed innovative techniques to be developed. Inoculative release techniques were established, including preparation before release, appropriate release time, release rate and special measures. Through experimental results and demonstrations, populations of aphelinid parasitoids and cecidomyid predators were able to establish and play very important roles in pest control on tomato, cucumber, and ornamental crops grown in greenhouses. Parasitism of the whiteflies, *Trialeurodes vaporariorum* and *Bemisia tabasi* was as high as 85% to 96%. Natural enemies released also effectively suppressed aphid populations on tomato and cabbage crops. Egg parasitism of the cabbage butterfly, *Pieris rapae*, and the cotton bollworm, *Helicoverpa armigera*, by *Trichogramma* wasps reached 78% to 95% on average. It was shown that natural enemies can suppress populations of target insect pests to below the economic threshold in greenhouse vegetable crops. When these techniques are combined with other non-chemical means of control for diseases and non-target insect pests, such as application of target specific fertilizers, augmentative biological control practices could greatly reduce the utilization of chemical pesticides, making non chemically-polluted vegetable products possible. A great economic benefit was acheived in 11,000 ha of biological control demonstration areas in Hebei, Beijing and Tianjin, by implementing the above augmentation biocontrol techniques from 2001 to 2004.

INTRODUCTION

As the most important method of vegetable production, greenhouses are becoming more and more prevalent in North China, and people are paying more attention to greenhouse pests.

Controlling greenhouse pests using chemical pesticides raises environmental concerns and can result in problems such as the development of resistance in pests. The use of biological control can overcome these problems while still providing adequate pest control.

ARTHROPOD PESTS AND THEIR NATURAL ENEMIES IN GREENHOUSES

The main arthropods that are greenhouse pests in North China are the greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood), tobacco whitefly, *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae), cabbage aphid, *Brevicoryne brassicae* (L.) (Hemiptera: Aphididae), and several acarid species. There are also other pests in greenhouse, such as *Tetranychus urticae* Koch (Hemiptera: Tetranychidae), *Polyphagotarsonemus latus* Banks (Hemiptera: Hemisarcoptidae), *Liriomyza sativae* Blanchard (Diptera: Agromyzidae) and some coccids, etc. (Cheng 2002; He 1996; Qu *et al.* 2002; Shi *et al.* 1995; Zhang *et al.* 1997) These pests cause significant damage on the vegetables produced in these greenhouses.

There are many species of parasitic wasps that attack whitefly, including 34 from the genus *Encarsia*, 14 of the genus *Eretmocerus*, and several species of *Amitus* and *Metaphycus*. In China there are about 19 species of parasitic wasps which include *Encarsia formosa* Gahan, *Encarsia pergandiella* Howard and *Eretmocerus mundus* Mercet (Hymenoptera: Aphelinidae). Approximately 114 species (9 orders, 13 families) of whitefly predators are known to exist in China. Some of the most important of these are *Lygus pratensis* L. (Hemiptera: Miridae), *Chrysoperla sinica* Tjeder (Neuroptera: Chrysopidae) and several predatory mites (Zhang *et al.* 2003; 2004).

Some predators of greenhouse aphids were found to be: *Leis axyridis* Pallas, *Propylea japonica* Thunberg, *Coccinella septempunctata* L., *Adonia variegata* Coeze (Coleoptera: Coccinellidae), *Syrphus corollae* F., *Epistrophe balteata* De Geer, *Lasiopticus Pyrastri* L., *Sphaerophoria scripta* L. (Diptera: Syrphidae), *Aphidoletes apidimyza* Rondani (Diptera: Cecidomyiidae), *Eringonidium graminicolum* Sundevall (Araneae: Erigonidae), *Pardosa T-insignita* Boes *et* Str. (Araneae: Lycosidae), *Chrysopa sinica* Tjeder, *Chrysopa septempunctata* Wesmael, *Chrysopa formosa* Brauer (Neuroptera: Chrysopidae), *Hemerobius humuli* Linnaeus (Neuroptera: Hemerobiidae), *Nabis sinoferus* Hsiao, *Nabis stenoferus* Hsiao (Hemiptera: Nabidae), *Orius minutus* L. (Hemiptera: Anthocoridae), and *Deraeocoris punctulatus* Fall (Hemiptera: Miridae). Parasitoids that help control these greenhouse aphids include species from the hymenopteran families: Ichneumonidae, Braconidae, and Chalcidae. As well, a parasitic fungus (Chen 2002; Chinese Academy of Science (Zooscopy Institute) 1978; He *et al.* 1986; Liu 2000; Xia *et al.* 2004).

Non-parasitic natural enemies of phytophagous mites found in China include ladybird beetles, the anthocorid, *Phytoseiulus persimilis* Athias-Henriot (Acariformes: Phytoseiidae), and *Campylomma chinensis* Schuh (Hemiptera: Mirridae). It has been reported that *P. persimilis* successfully controls phytophagous mites both in its native habitat, and in other habitats abroad (Dong *et al.* 1986; Liang 2004; Yang *et al.* 1989).

Worldwide, arthropod natural enemies of thrips include species of Nabidae, Miridae, Anthocoridae, Sphecidae, Eulophidae, Trichogrammatidae, Mymaridae, Coccinellidae, Syrphidae, Dolichopodidae, Cecidomyiidae, Aeolothripidae, and some predatory mites (Ananthakrishnan 1973; Lewis 1973).

In China, there are few reports about the natural enemies of common thrips. Qing *et al.* (2004) found that predatory arthropods include *Campylomma chinensis*, *Cyrtorhinus lividipennis* Reuter (Hemiptera: Miridae), *Orius simillis* Zheng (Hemiptera: Anthocoridae), *Geocoris pollidipennis* F. (Hemiptera: Lygaeidae), *Scolothrips takahashii* Piesneer (Thysanoptera: Thripidae), some ladybird beetles, spiders, and ants. A total of 10 families and about 20 species of predators; among them, *C. chinensis* are the dominant natural enemies (Qing *et al.* 2004).

The known predatory arthropods of leaf miners include *Propylaea japonica*, *C. septempunctata*, *E. graminicolum*, and *P. T-insignita*. The parasitic wasps include *Opius spp.* and *Dacnusa spp.* (Lu *et al.* 2000); species of *Chrysocharis*, *Dacnusa*, *Diglyphus*, *Opius*, *Neochrysocharis*, *Hemiptarsenus* and *Halticoptera* are some of the more common parasitoids found to control leaf miner (Chen *et al.* 2001).

MASS-REARING OF BENEFICIALS IN CHINA

In recent years, techniques for mass-rearing beneficials have been developed and improved, to efficiently control major arthropod greenhouse pests. Several species can now be produced on a large-scale, and released in greenhouses in China. Beneficials such as *Trichogramma spp.*, *E. formosa*, *Eretmocerus spp.*, *P. persimilis* and *Aphidoletes apidimyza* have been successfully mass-produced by the Hengshui Tianyi Bio-control Company, Dryland Farming Institute.

TRICHOGRAMMA SPP.

In order to rear *Trichogramma spp.* with high selectivity to vegetable pests, *Sitotroga cerealla* eggs were used as host eggs. Several species, including *T. evanescens*, *T. pretisum*, *T. brassicae*, *T. embryophagum*, and *T. cacoaciae* can be mass-produced using this system. For mass-production of *S. cerealla* eggs, new production line and rearing techniques were developed. A specially made egg auto-collection machines were used and over 10 million eggs could be collected in 24 hours, provided there is an ample supply of emerged moths. Other equipment for use in moth rearing and egg purification was also developed by Hengshui Tianyi Bio-control Company in Hebei, China (Zheng 2003; 2004).

ENCARSIA FORMOSA AND *ERETMOCERUS* SP.

It is very important to find a proper variety of fod plants to feed to the insect hosts of both *Encarsia* and *Eretmocerus*. Since tobacco can be perennially cultured in greenhouses, varieties of tobacco were screened for their suitability as host plants for whitefly. Selection of these varieties ensures that sufficient numbers of whiteflies survive for a longer time, offering ample host accessibility to both *Encarsia* and *Eretmocerus*. Wasps oviposit into the young whitefly larvae, and when they develop to their pupal stage they are harvested. A special mass-production procedure of *Encarsia* and *Eretmocerus* has been developed by HTBC in Hebei, China (Zheng 2004).

APHIDOLETES APIDIMYZA

For mass rearing of *A. apidimyza*, insect hosts and their host plants were selected. The HTBC has also developed mass-rearing techniques of *A. Apidimyza* (Zheng 2004).

OTHER BENEFICIALS

Jiexian Jiang studied the mass-rearing and application of *Aphidius gifuensis*, and found that this parasitoid could be used to control the damage caused by aphids (Jiang *et al.* 2003). Although there are many natural enemies of aphids worldwide, only *A. apidimyza* has been reared on a large scale, and used in greenhouses.

It is very difficult to mass-rear ladybird beetles with artificial food. It has been reported however, that an artificial food diet, suitable for a female ladybird beetle to lay eggs on, has been successfully produced in China. An artificial diet for lacewings has also been successfully made, what's more, all stages of lacewing could develop on artificial eggs.

RELEASE OF BENEFICIALS AND BIO-CONTROL IN GREENHOUSES

PREPARATION BEFORE RELEASE

To satisfy the need for a controlled effect, some preparatory measures need to be taken before the release of natural enemies. These measures include: growing clean seedlings for transplanting, cleaning and sterilizing greenhouses for about 15 days and fixing screens on ventilation devices to prevent access by outside insects. The above precautions allow inoculative releases of beneficials to be successfully made after transplanting seedlings into greenhouses.

RELEASE OF *ENCARSIA FORMOSA* TO CONTROL WHITEFLY

These tiny wasps lay eggs inside the scales of developing whitefly larvae. The parasitoids then complete their development inside the whitefly larvae, killing the host in the process. Upon emergence, adults immediately begin to search for other larvae. Parasitized whitefly larvae are easy to recognize, as they will turn black over time.

When the average number of adult whitefly reaches 1000 in one greenhouse (about 0.05ha.), it is time to release *E. formosa*. The ratio of enemy versus adult pests is 3:1 (3000-5000 wasps per house). Wasps are introduced every 7-10 days, and after 3-4 releases, a balance is reached between wasps and whiteflies, and the introduction of the parasitoids to the greenhouse can be stopped. The temperature of the greenhouse containing the wasps should be controlled and maintained between 15-35 °C.

RELEASE OF *APHIDOLETES APIDIMYZA* TO CONTROL APHIDS

To control aphids successfully, *A. apidimyza* was introduced into the greenhouse before the aphid could damage the vegetables. These predators cripple the aphids by quickly injecting a paralyzing toxin, then sucking out the body fluid, leaving a shriveled aphid husk still attached to the leaf. When aphid numbers are high, they may kill many more aphids than they eat. Fully-grown predator larvae leave the plant to pupate in the soil.

If some wheat plants containing wheat aphid are brought into the greenhouse, *A. apidimyza* will survive on these aphids and the aphids will not feed on the greenhouse vegetables. As a result, initial aphid numbers can be controlled at low-density levels. At the first occurrence of aphids, *A. apidimyza* was released in the ratio of 1 larvae for every 20 aphids, and had a controlling effect after 2-3 continual releases.

RELEASE OF *TRICHOGRAMMA* SPP. TO CONTROL PESTS OF LEPIDOPTERA

In greenhouses without screen or ventilation, pests of *Lepidoptera* may seriously damage vegetables. In this case, *Trichogramma* spp. should be introduced. Several days after their introduction into the greenhouse, *Trichogramma* spp. wasps will emerge from parasitized eggs and seek out a new lepidopteran host.

RELEASE OF *PHYTOSEIULUS PERSIMILIS* TO CONTROL PHYTOPHAGOUS MITES

The predatory mite, *P. persimilis*, is a very good natural enemy to control phytophagous mites. To efficiently control these mites, the ratio between *P. persimilis* and phytophagous mites should be about 1:10 to 1:20. *P. persimilis* was released every 7-10 days, and after 3-4 weeks the number of phytophagy mites dropped notably (Dong *et al.* 1986; Li *et al.* 2004).

This predator does not feed on the plant or shrub and is fully dependent on the spider mite and its eggs for food. Generally, only one introduction of *P. persimilis* is required each season, because the predator population remains in low numbers once control is gained. To obtain optimal reproduction rates, the temperature of the greenhouse should be maintained between 21-27°C.

OTHER BIOLOGICAL CONTROL METHODS IN GREENHOUSES

PATHOGENIC FUNGI OF INSECT PESTS

Most pathogenic fungi used for the control of whitefly are Hyphomycetes including species of *Paecilomyces*, *Verticillium*, and *Aschersonia*. *Aschersonia aleyrodis* Webber (Sphaeropsidales: Sphaeriodaceae) is an important pathogenic fungus of whitefly and coccids, and much attention was given to *Paecilomyces fumosoroseus* Wize and *Verticillium lecanii* Zimmermann (Moniliales: Moniliaceae) (Xiao 2002; Zhang 2003; 2004).

There are 37 species of pathogenic fungus that can be used for the control of aphids. These are included within 9 genera of Entomophthorale and 7 genera of Hyphomycetes; among these, *Beauveria bassiana* Balsamo (Moniliales: Moniliaceae) and *V. lecanii* can also be used to control common thrips (Li *et al.* 2005; Qin *et al.* 2001).

BIOLOGICAL PESTICIDES

The main biological pesticides used in greenhouses today include *Bacillus thuringiensis* (Berliner) [*Bt*], abamectin, Azadirachtin and Polynactin. Athough pheromones were used to control pests during the 1960's, there are few reports on this topic. Due to the closed conditions within a greenhouse environment, kairomones that are produced by insect pests are not useful to many natural enemies. Some plants can produce metabolites such as terpene, alkene, alkaloid, lignin, steroid, flavone and polysaccharide, which can then be used to control greenhouse pests. Naturally occuring pesticides such as plecocidin, which is developed from plants, can be used in greenhouses to control pests and will not lead to environmental problems.

YELLOW BOARDS

The use of yellow boards within a greenhouse environment can efficiently monitor the effects of biological control efforts. Approximately 20 yellow boards are sufficient in one house, and when hung properly in greenhouses, can attract whiteflies, aphids and leaf miners.

GREENHOUSE CONDITIONS IN CHINA

Currently, the total greenhouse area in China is over 2 million ha. These all fall within three different categories:

Glasshouse. The glasshouse is the style of greenhouse that provides the optimal conditions for use with natural enemies. There are about 1300ha of glasshouse in China, making up no more than 0.1% of the total greenhouse area. The main advantage to using this type of greenhouse is the control one has over the environmental conditions through the use of heaters, fans and other devices. The temperature can be maintained above 15°C during the cold season and below 35°C during the hot season, and the humidity in these glasshouses can also be reduced or raised to an optimal level. Optimal control can easily be reached after the release of the beneficials into the glasshouse; however, much attention should be paid to monitoring the development of the insect pests while different crops with different growing seasons are harvested in the same house.

Cold plastic house. One of the most extensively used greenhouse styles in China is the cold plastic house. These are covered only by plasticand crops cannot be grown during the wintertime; instead crops are produced during two growing seasons. For the first season, crops are planted in spring and harvested in summer. Since pests are not a serious problem in spring, farmers usually neglect to control them at the beginning of planting. Farmers also pay little attention to the pests in the summer, since the vegetables are beginning to be harvested. For the second season, crops are planted in summer or the beginning of autumn, and it is at this time when high populations of pests occur. Most farmers grow seedlings without using effective pest prevention methods; as a result, many pests are easily transported from outside into the plastic house when vegetables are transplanted. These high populations of pests make control much more difficult when releasing natural enemies.

Warm plastic house. Another style of greenhouse, used most extensively in China, is the warm plastic house. A thick wall built on the north side of the house prevents penetration of the strong wind during cold winters, and allows crops to grow year-round. During most of the year, throughout each growing season temperature and humidity are satisfactory to release beneficial arthropods. It is only during the wintertime, because there is generally no heating temperature and humidity levels are unfavourable and the use of natural enemies is not possible.

In China, most of the greenhouses used are made of plastic, and are either a warm house or a cold house. To obtain efficient control of arthropod pests after the release of beneficials, we strongly suggest that farmers grow clean seedlings and use screen on the ventilation systems of their greenhouse, before applying biological control techniques.

With the improving demand for green food and the increasing greenhouse area, bio-control in greenhouses will have a more important place with regards to pest control and safe-food production. Improving bio-control and rearing measures will provide more efficient control over greenhouse pests.

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