VG106
An evaluation of egg parasitoids for the management of heliothis

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Know-how for Horticulture™

VG106

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SUMMARY

Heliothis (*Helicoverpa armigera* and *H. punctigera*) are serious insect pest of vegetable crops in Australia. Control has been achieved primarily through the use of chemical insecticides. However, the development of resistance, the increasing cost of developing new insecticides, and growing community concern over the effects of insecticides on the environment require that alternatives to this form of management be found. Biological control using egg parasitoids is one such method.

Egg parasitoids (*Trichogramma* and relatives) are small wasps (0.3-0.5 mm long) that attack the egg stage of moths, killing them before they hatch and cause damage. These wasps occur naturally throughout Australia, but are usually in insufficient numbers to adequately manage insect pests in agroecosystems. There use as biological control agents involves producing large numbers of individuals and releasing them into crops where there are pest eggs. The wasps then locate and attack pest eggs in the field. This process is known as inundation, and the wasp releases are called inundative releases. The project involved evaluating native egg parasitoids as inundative biological control agents of heliothis on lettuce, tomatoes and sweet corn.

Techniques for mass rearing egg parasitoids on eggs of the Angoumois Grain Moth, *Sitotroga cerealella*, were developed and passed on to three commercial companies in eastern Australia. *Sitotroga* were reared on whole wheat. Adult moths were collected and placed in automated mesh tumblers. Eggs were harvested from the tumblers daily and could be used to mass rear wasps. The wasps produced on grain moth eggs were then available for use in field trials against heliothis. Field trials were completed on sweet corn and tomatoes.

The most encouraging results were found on maize/sweet corn. A release of *Trichogrammatoidea bactrae* (420,000 females) made in a 0.25 ha fraction of a 40 ha maize planting at Dalby increased heliothis egg parasitism from 6.0% to 50.8% in three days, demonstrating that inundative releases of wasps can have a rapid impact on pest mortality.

Consistently high levels of egg parasitism in a sweet corn trial at Mulgowie resulted in 93.3% of cobs being marketable, indicating that egg parasitoids can successfully manage heliothis on sweet corn. The source of the parasitoids was, however, unclear. It seemed likely that natural populations of wasps were abundant in the region and contributed greatly to the levels of heliothis egg parasitism recorded. The value of inundative releases in such a situation needs clarifying. Additional research is required to determine the number of wasps (be they natural or released) necessary to achieve adequate pest control.

The results of inundative releases of egg parasitoids against heliothis in tomatoes at Bundaberg were disappointing. *Trichogramma ivelae* (eight releases) and *T. funiculatum* (one release) were released at approximately 300,000 females/ha and failed to manage a heliothis infestation. While the levels of egg parasitism became high towards the end of the crop (up to 87.7%), it was too late to have an impact on the severe heliothis infestation and no fruit was harvested. Again, data suggested that there was a natural population of wasps active in the region. The natural wasp population contributed to the levels of parasitism recorded in the release site, and in a nearby non-release site where parasitism peaked at 90.1%. The inundative releases appeared to have little immediate impact on heliothis mortality, indicating that *T.ivelae* may not have been the most suitable species for use against heliothis on tomatoes.

Glasshouse trials comparing the number of heliothis eggs parasitised by six native species of egg parasitoids found that *T. funiculatum* parasitised more eggs than other species on lettuce and tomatoes. This species has been provided to the commercial producers of *Trichogramma* in eastern Australia and should be available for use during 1994/95.

The inconsistent performance of wasps in field trials suggested that they may be better used as tools in an Integrated Pest Management (IPM) program. It was therefore necessary to determine if some insecticides were compatible with egg parasitoids. The effects of residual chemical on the survival of two species of egg parasitoids were assessed in the laboratory. Those chemicals and fungicides commonly used on lettuce were tested. There was no significant difference between survivorship on leaves sprayed with *Bacillus thuringiensis* (Dipel) or chlorfluazuron (Helix) when compared to a control sprayed with water, suggesting that these insecticides are compatible with *Trichogramma*. Survivorship on leaves sprayed with endosulfan, permethrin and methomyl was not high. Further research is required to determine if theses chemicals can be used in IPM programs with *Trichogramma*. The fungicides metalaxyl + mancozeb (Ridomil) and procymidone (Sumisclex) did not adversely affect wasp survival, and are therefore suitable for use with egg parasitoids.

The research found that native egg parasitoids have potential at managing heliothis, but their value as inundative biological control agents remains unclear. Further research is necessary to understand the impact of inundative releases on heliothis egg mortality, and to also clarify the role of natural wasp populations in field situations. Releases of egg parasitoids may best contribute to heliothis management when used in IPM programs.

THE PROBLEM

Heliothis, *Helicoverpa armigera* (Hübner) and *H. punctigera* (Wallengren) (LEPIDOPTERA: NOCTUIDAE), are serious pests of field, horticultural and ornamental crops in Australia (Zalucki *et al.*, 1986). The annual loss to the economy (in terms of crop loss and pest management costs) caused by heliothis in Queensland averaged $73 million (range $23-162 million) in 1989, including $ 6 million for tomatoes, sweet corn and lettuce (McGahan *et al.*, 1991).

INDUSTRY SIGNIFICANCE - THE INSECTICIDE DILEMMA

To date, insecticides are the most widespread commercially used means of controlling heliothis on vegetables in Australia. The regular application of insecticides provided reliable control of heliothis until the early 1970's when resistance to DDT by *H. armigera* was first detected (Twine and Kay, 1973; Wilson, 1974).

Resistance to the synthetic pyrethroids was reported at Emerald in 1983 (Anon., 1983), and is now widespread in the major cropping areas of eastern Australia (Daly, 1988).

Heliothis resistance to chemicals in lettuce has been a problem on the Darling Downs in recent times. Larvae collected from Cambooya displayed 100% resistance to synthetic pyrethroids, 50% resistance to endosulfan, 50% resistance to methomyl and 13% resistance to thiodicarb (Robyn Gunning, pers. comm.). Resistance to thiodicarb has also been a problem to sweet corn growers.

There are few cost effective insecticides on the horizon, with the cost of developing a new insecticide estimated at approximately $US50 million (Forrester, 1989).

The combined effects of insecticide resistance, legislation, increased public concern, and higher insecticide costs suggests that insecticides will not remain the sole pest management tool for the horticultural industry in the future. Supplementary control tactics must therefore be developed and integrated into existing management strategies.

Numerous natural enemies have been reported to attack heliothis. Waterhouse and Norris (1987) list 45 species of dipteran and hymenopteran parasites reared from heliothis in Australia. Of these, the egg parasitoids are the easiest to rear, store and transport, and trials with these natural enemies against heliothis have been undertaken in Australian horticulture in the past (McLaren and Rye, 1981, 1983).

EGG PARASITOIDS AS BIOLOGICAL CONTROL AGENTS

Egg parasitoids of the genus *Trichogramma* are frequently used as inundative biocontrol agents of lepidopterous pests (Stinner, 1977). Inundative biological control involves releasing large numbers of natural enemies to overwhelm a pest population and maintain pest density below

that level causing economic damage (Stehr, 1982). Egg parasitoids have an advantage over larval parasitoids in that they kill pest insects before they can cause damage.

Trichogramma and relatives are known as parasitoids and differ from true parasites in that: a) the development of an individual destroys its host, b) they are parasitic as larvae only - the adults are free living, and c) their action on a host population more closely resembles that of a predator than a parasite (Doutt, 1959). *Trichogramma* are called as egg parasitoids because they attack the egg stage of their hosts, killing them before they hatch.

The female *Trichogramma* inserts her ovipositor through the shell of a host egg and lays her eggs inside the host egg (Figure 1). The egg hatches and the wasp larvae consume the contents of the host egg as they develop. Metamorphosis occurs inside the host egg. The larvae pupate, eventually emerging as adult wasps. The entire life-cycle, from egg to wasp, takes approximately ten days at 25°C. The adult wasps feed on nectar and exudate from the puncture sites of parasitised host eggs and live for approximately five days.

Host egg colour can be useful when assessing levels of parasitism. A heliothis egg that has been parasitised by a *Trichogramma* wasp turns black, having a uniform black shell, after four days at 25°C. The colour of unparasitised eggs changes with age also. One, two and three-day-old unparasitised eggs are white, brown and 'black' respectively. The 'black' colour of the three-day-old eggs is due to the black head capsule of the developing larva that is visible through the clear egg shell (the egg shell of a parasitised egg is black).

The Cotton Experience

There has been, and continues to be, enormous interest in evaluating *Trichogramma* for heliothis management in cotton, particularly in the U.S.A.. The American research suggests that *Trichogramma* have little potential for commercial acceptance. King and Coleman (1989) state that 'there is limited experimental evidence to support the commercial use of *Trichogramma* for *Heliothis* (*Helicoverpa*) control'.

Some workers (Stinner *et al.*, 1974; Ables *et al.*, 1979) have reported a reduction in heliothis larval numbers following *Trichogramma pretiosum* Riley releases; while others (King *et al.*, 1986) have reported an increase in egg parasitism but no reduction of larval numbers. All workers have found that inundative *Trichogramma* releases can increase egg parasitism levels, but none have established that such releases can maintain heliothis densities below those that will prevent economic damage in cotton.

The apprehensions that research workers have about the use of *Trichogramma* for heliothis control in cotton can perhaps be attributed to the inconsistent effectiveness of the parasitoids in field trials. Despite this, it is clear from the reported widespread commercial use of *Trichogramma* to manage heliothis in cotton - over 0.5 million hectares in the USA and South America alone (Hassan, 1984) - that egg parasitoids have some potential as biological control agents.

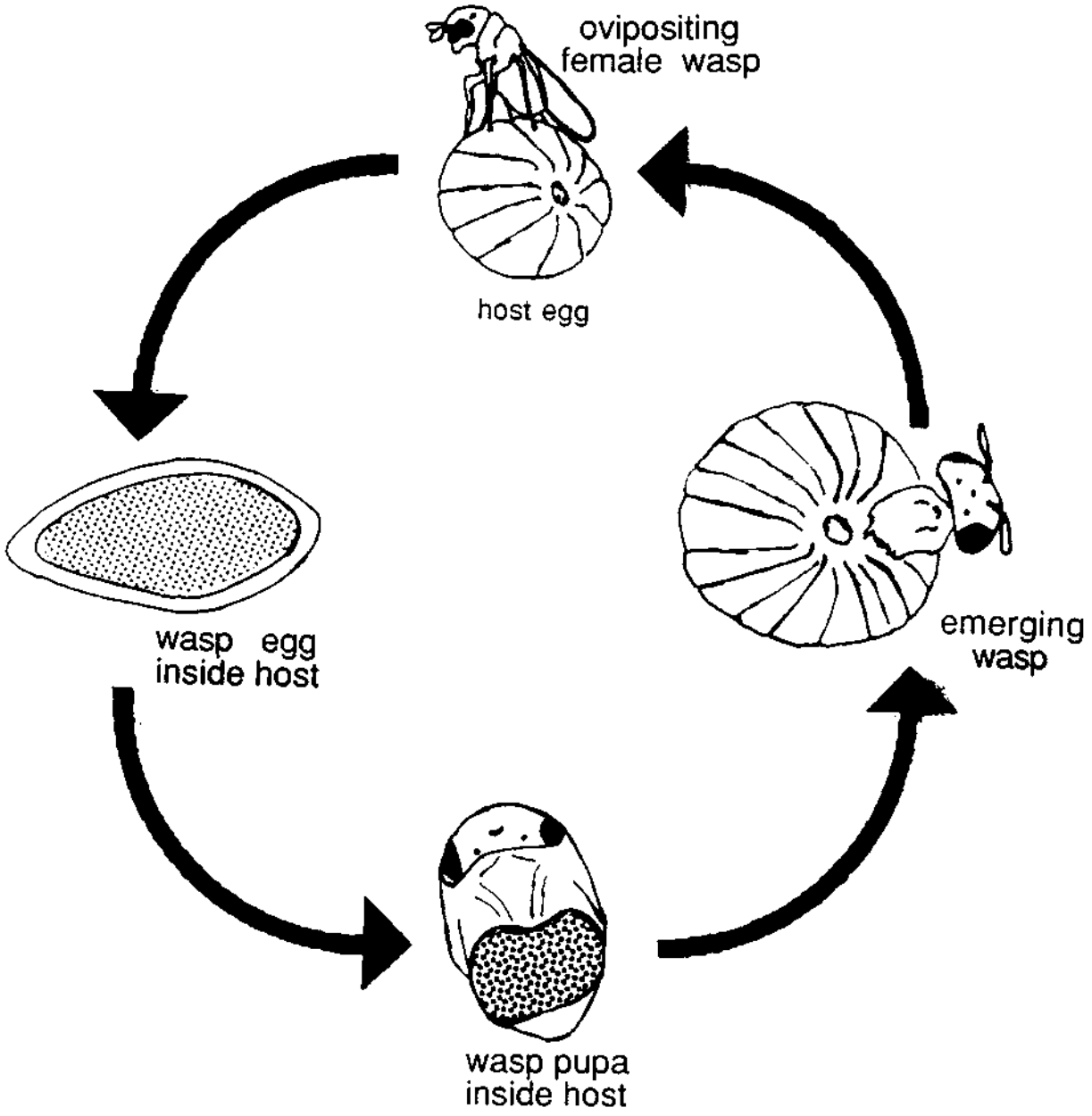


FIGURE 1: Generalised life-cycle of an egg parasitoid.

In contrast to the cotton experience, the use of *Trichogramma* to manage other insect pests in a range of crops throughout the world is commercially accepted. *Trichogramma* have been successfully used to control the European corn borer, *Ostrinia nubilalis* (Hübner), in 34,000 ha of maize in Europe (Hassan, 1984); and sugar cane, rice and forest pests in China and the USSR (Hassan, 1984; Cock, 1985). Hassan (1984) reports that *Trichogramma* are commercially used to manage pests in over 18.5 million hectares of crop throughout the world. The largest areas treated with *Trichogramma* are in the USSR (16 million hectares) and China (1.2 million hectares). *Trichogramma* also display promise for pest management in tomatoes (McLaren and Rye, 1983; Oatman *et al.*, 1983b), avocadoes (Oatman and Platner, 1985) and sweet corn (Neil and Specht, 1990).

TRICHOGRAMMA USE IN VEGETABLES

Overseas

Trichogramma are reputedly used to successfully manage European corn borer in sweet corn in Germany (Neuffer, 1982; Hassan, 1982), USSR (Khloptseva, 1991), Switzerland (Bigler, 1986; Bigler and Brunetti, 1986), and Bulgaria (Karadjov, 1989).

The use of *Trichogramma* against heliothis in sweet corn has given mixed results. Early workers found that heliothis could not be managed by *Trichogramma* (Fletcher, 1935; Larrimer, 1935). However, research interest in *Trichogramma* persisted because natural populations of egg parasitoids were found to have an impact on heliothis in unsprayed crops (Oatman, 1966; Oatman *et al.*, 1983a; Meierrose and Araujo, 1986; Hoffmann *et al.*, 1990).

Neil and Specht (1990) released 275,000-366,000 *T. pretiosum* per hectare against heliothis in sweet corn in Nova Scotia. They found that larval populations were reduced up to 74% in *Trichogramma* release plots, compared with reductions of up to 100% in insecticide plots. These results were encouraging, especially when no attempt was made to use native or selective strains of *Trichogramma* that may have been better adapted to climatic conditions (the wasps used in the trial came from Texas, U.S.A.).

Trichogramma have also displayed promise at managing heliothis in tomatoes (Oatman and Platner, 1971; Oatman and Platner, 1978). Yadav *et al.* (1985) found that *Trichogramma chilonis* Ishii was successful at managing heliothis in Indian tomatoes. When wasps were released at a rate of 250,000/ha every week (for 10 weeks) there was up to 75% reduction in fruit damage and 96% egg parasitism.

Trichogramma can also be important in IPM programs. Oatman et. al. (1983b) found that weekly application of *Bacillus thuringiensis* (*B.t.*) and bi-weekly applications of *Trichogramma* (56,000/ha per release) reduced fruit damage in fresh market tomatoes in California. Total fruit injured was 0.7, 1.6 and 7.1% in commercial (chemical), IPM (*B.t.* and *Trichogramma*) and control plots respectively. Similarly, Trumble and Alvarado-Rodriguez (1993) found that an IPM program for fresh market tomatoes in Mexico

utilising 4-9 releases of *T. pretiosum* (200,000/ha) against heliothis was more profitable than conventional (chemical) control programs. Mean fruit damage in spring planted crop managed by IPM was not significantly different from that in conventional crop.

It is clear that *Trichogramma* have potential to be used as biological control agents of heliothis, although the commercial acceptance of them as a form of pest management has been adopted slowly.

Australia

In Australia there has been little research evaluating egg parasitoids for heliothis management. Research in the 1970's was on cotton and involved inundative releases of the exotic wasp *T. pretiosum* (Woods, 1981; Twine and Lloyd, 1982). No attempt was made to evaluate native species as biocontrol agents. However, the research that was carried out in tomatoes in the early 1980's involved inundative releases of the native egg parasitoid *Trichogramma ivelae* Pang and Chen.

McLaren and Rye (1983) reported significant reductions in tomato fruit damage after weekly releases of *T. ivelae* (100,000/ha). In the release sites, damage at the first harvest ranged from 0.2-28.9% infested fruit, a reduction of 98% and 55% respectively. It was concluded that the 'inundative release of *T. ivelae* for control of *H. punctigera* had commercial potential' (McLaren and Rye, 1983).

The research presented here evaluates native egg parasitoids as biological control agents of heliothis on lettuce, tomatoes and sweet corn. It is hoped that the use of egg parasitoids will contribute towards the development of sustainable crop production practices in the future.

RESEARCH OBJECTIVES

* To sample natural egg parasitoid populations.
* To establish laboratory colonies of different species of egg parasitoids.
* To identify those species that have potential as biological control agents.
* To evaluate the effects of insecticides on the survival of adult parasitoids.
* To develop and maintain a facility for mass rearing wasp egg parasitoids.
* To evaluate inundative releases of wasps as a heliothis management tool.

INSECT REARING

A substantial component of this project involved rearing insects, primarily heliothis (*Helicoverpa armigera* (Hübner)), wasp egg parasitoids (*Trichogramma* species and *Trichogrammatoidea* species), and the Angoumois grain moth (AGM), *Sitotroga cerealella* (Olivier). Heliothis eggs were required to conduct laboratory and glasshouse studies evaluating the potential of wasps to destroy eggs. Some cultures of wasp parasitoids were also continually maintained on heliothis eggs.

Large numbers of *Trichogramma* were required for field release programs. Consequently large numbers of insect eggs were needed to rear the wasps. Heliothis are too difficult and expensive to rear in sufficient numbers to mass rear wasps. Therefore a factitious host was used that was comparatively easier, and inexpensive, to mass rear than the target host (heliothis). Two factitious host insects are commonly used in western countries to mass rear *Trichogramma*, viz. the Mediterranean flour moth, *Ephestia kuehniella* Keller, and AGM (*S. cerealella*). The AGM was used in this study.

Heliothis (Helicoverpa armigera)
Heliothis were reared on a navy bean based diet which was modified from that described by Teakle and Jensen (1985).

Larval Diet Composition

a.	navy bean		258 g
	tap water		1410 ml
b.	agar		37.5 g
	tap water		909 ml
c. dry mix:	wheat germ		180 g
	torula yeast		150 g
	l-ascorbic acid		9 g
	nipagin		9 g
	sorbic acid		3 g
d.	formaldehyde (40%)		3 ml
e.	mould inhibitor		7.8 ml
	mould inhibitor preparation:		
	propionic acid	42 ml	
	phosphoric acid	4 ml	
	water	54 ml	

Moth Diet Composition

water	1000 ml
sugar	100 g
l-ascorbic acid	6 g
terramycin soluble powder	2 g

The navy bean/water (a) and agar/water (b) mixtures were steamed separately in covered beakers for two hours. Both mixtures were then cooled for 5-10 minutes to approximately 70°C. The beans, dry mix (c) and formaldehyde (d) were mixed in a waring blender. The agar and mould inhibitor (e) were then added and remixed.

The diet was poured into stainless steel larval rearing trays measuring 300x300x20 mm. Each tray was lined with plastic film before the diet was poured. This aided cleaning at the completion of rearing. The diet was allowed to cool at room temperature. An aluminium grid was then pressed into the diet to produce 196 square cells (20x20 mm) that were 20 mm deep. Separate rearing cells are required when rearing heliothis because the larvae are cannibalistic (Twine, 1971).

Loose heliothis eggs were dispensed onto the diet by inverting a plastic vial with a stainless steel gauze lid (8 strands/cm; 28 SWG; 0.9 mm aperture) over the tray and sprinkling the eggs onto the diet. Each tray was then covered with perforated aluminium sheeting (0.81 mm holes; 0.6 mm thick; 31% open area) to contain larvae to cells and prevent cannibalism or escape. Two large rubber bands held each perforated sheet in position. The covered trays were wrapped in brown paper and held in a constant temperature room (approximately 25°C and 70% R.H.) for 24 days until pupation (Figure 2a).

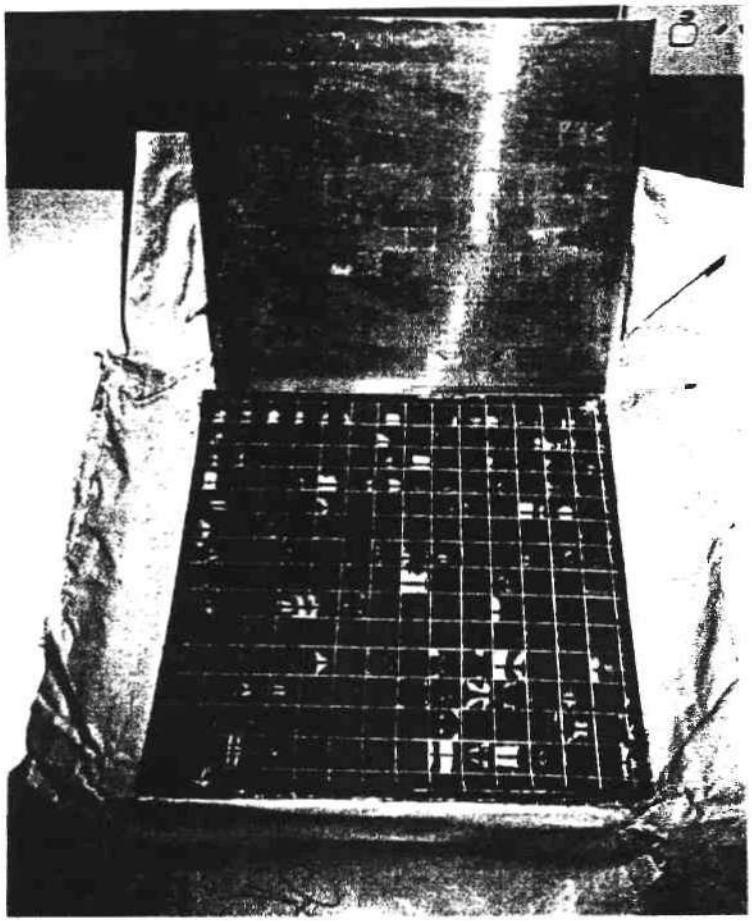
The pupae were removed from the diet trays with feather forceps and washed in a sodium hypochlorite solution (0.167% a.i.) for three minutes and then rinsed in water. They were then placed in an aluminium tray with vermiculite, and the tray was placed in a mating cage (300x300x300 mm) covered with fine nylon cloth. A plastic container of sucrose solution (moth diet) was provided for emerging moths. Two cotton dental wicks were passed through the lid of each moth diet container and acted as feeding stations for the moths.

Moths emerged approximately 7 days after pupation and were allowed to mate in the mating cage for two days. The moths were then transferred to aluminium oviposition cages (Figure 2b) (after Teakle and Jensen, 1985). Diet containers were also placed in each oviposition cage. The moths laid eggs on the paper towelling walls of the cages. The paper walls (containing the eggs) were removed daily. The eggs could be stored at 8±1°C for 4-5 days and maintain reasonably high levels of hatchability, or could be stored for longer periods (up to two weeks) if they were used for rearing egg parasitoids.

The eggs were removed from the paper towelling by gently washing them in a 0.167% a.i. sodium hypochlorite solution for three minutes and rinsing them in water. The eggs were poured into a plastic squeeze bottle and dispensed onto pieces of cotton cloth that were placed on flyscreen frames. These frames were stacked in a rack and the eggs were allowed to dry at room temperature (approximately three hours). The eggs were removed from the cotton cloth by brushing them in short, gentle strokes with a soft paint brush. They were then ready for dispensing onto larval diet trays.

Sitotroga cerealella (AGM)

An experimental facility for rearing egg parasitoids on AGM eggs was developed at QDPI Toowoomba. AGM larvae were reared on wheat using techniques adopted from those described by others (Morrison, 1985b; Laing and Eden, 1990).



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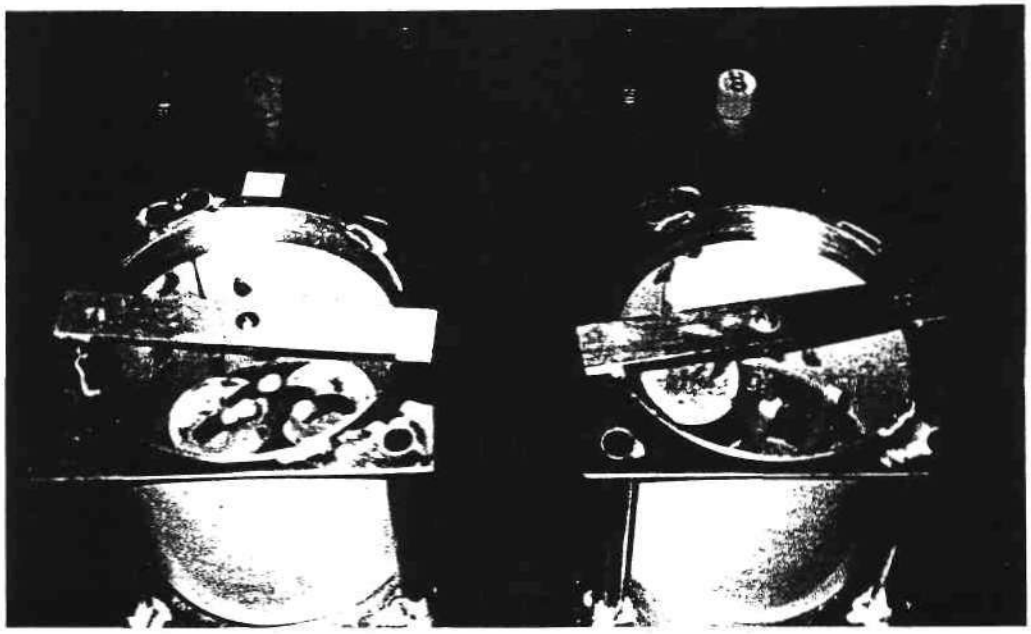

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FIGURE 2: Some of the equipment used to rear heliothis: (**A**) Larval rearing trays showing pupae. (**B**) Moth oviposition cages.

Chemically free wheat was used to rear AGM larvae. All wheat was heated in an incubator at 65°C for 24 hours before use to kill any insects or mites that may have been in the grain. The sterilised grain was treated with a miticide (dicofol - 0.113% a.i.) at a rate of 600 ml per 5 kg of wheat to prevent infestation by mite pests. The treated grain was then placed in a sealed plastic container and held in a refrigerator (approx. 4°C) for three days to restore the grain to a favourable moisture content of approximately 15% (Morrison, 1985b).

Rectangular AGM larval rearing cribs (1200x600x20 mm) were constructed of 19x19 mm timber framing on two sides and a base. Three timber dividers (580 mm long) divided each crib into four compartments 275 mm wide. Raised aluminium mesh (Expamet 601A) was stapled either side of the timber frame and 2.5 kg of treated wheat was poured into the top of each compartment. Two timber spacers (30 mm long) were stapled in each compartment to maintain the wheat at a uniform thickness of approximately 20 mm throughout the crib.

The cribs were held in units to collect emerging moths. Each moth unit consisted of a base funnel, and one or two sets of hangers above (Figure 3a). The base was made from galvanised sheet metal (24 gauge) and consisted of a rectangular opening (1320x710 mm) tapering down to a cylinder 70 mm high and 186 mm in diameter. Two metal lugs protruded from the cylinder that permitted a 4 L plastic container to be attached to the bottom of the funnel. Two holes (120x60 mm) were cut into the plastic containers and covered with fine stainless steel gauze (24 strands/cm; 37.5 SWG; 0.25 mm aperture) to provide ventilation. The base was supported on a steel frame measuring 1320x710x800 mm, with castors on each leg so that the unit could be moved easily for maintenance.

The hangers that held the cribs consisted of five 'U' shaped galvanised sheet metal troughs (1320x40x25 mm) riveted to the bottom of a steel frame (1320x710x635 mm). These troughs acted as guides for the cribs which were slid into the hangers and held at approximately 75° by 'L' shaped galvanised sheet metal support brackets at the top of the steel frame. The guides were 100 mm apart. A second hanger constructed in the same manner could be bolted to the top of the first hanger so that ten cribs could be stored in the one unit. A fine nylon cover attached with velcro enclosed the AGM moths emerging from the grain.

The treated grain was infested with enough AGM eggs to produce two eggs per kernel of wheat. This required approximately 1.2 g of AGM eggs per kilogram of wheat (there were 28,300 kernels of wheat/kg, and 50,400 AGM eggs/g), or 12 g AGM eggs per 10 kg crib (3 g eggs per compartment). Cribs containing newly treated wheat were held horizontally in steel storage racks for infesting with AGM eggs. The wheat was misted with water from a hand held sprayer, and the eggs were sprinkled onto the wheat by shaking them from a small plastic bottle with a stainless steel gauze lid (8 strands/cm; 28 SWG; 0.9 m aperture).

AGM eggs could be stored at 5±1°C (50±5% R.H.) for up to five days and still maintain high levels of egg hatch (> 75%). After this, egg hatchability declined rapidly.

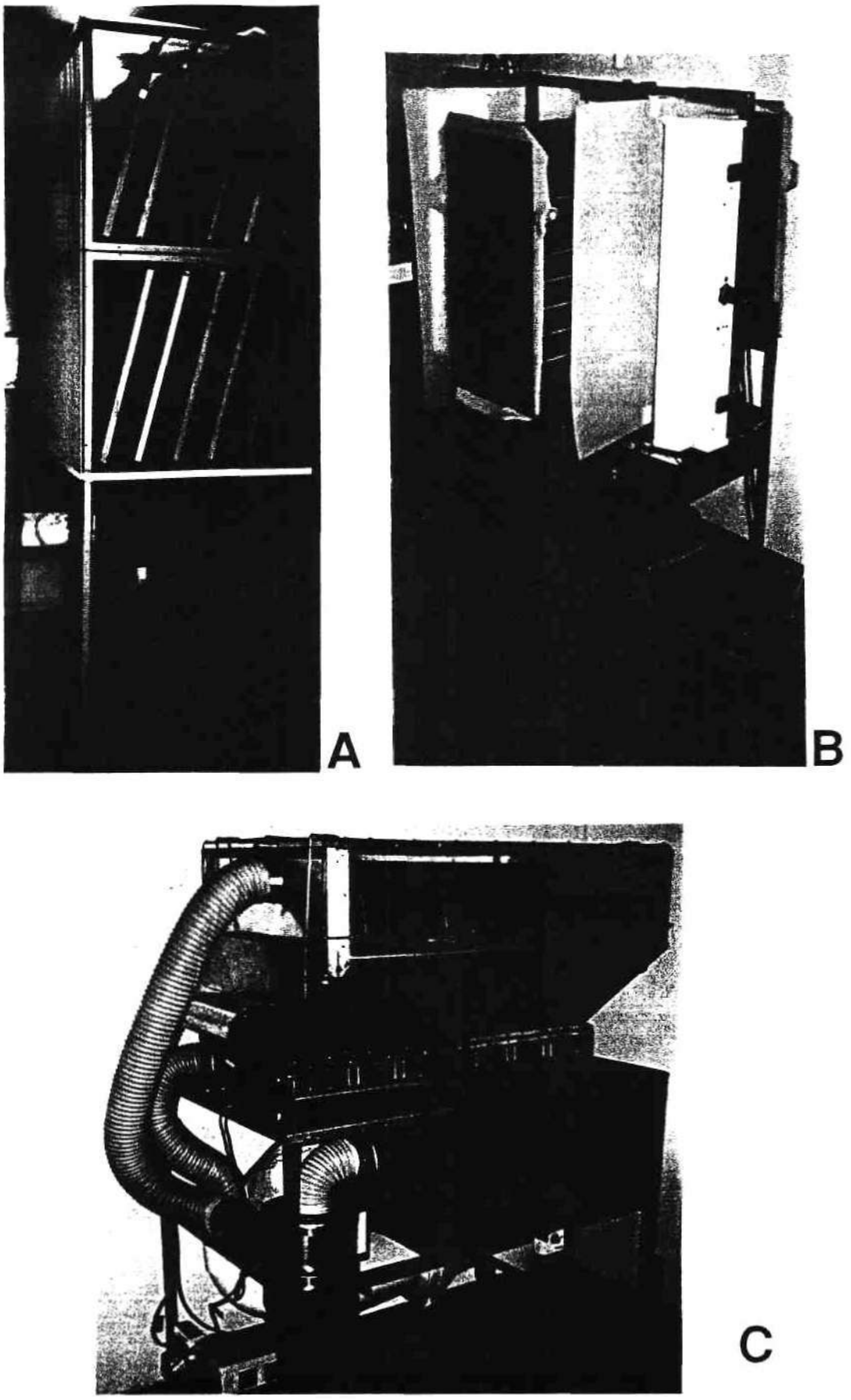

FIGURE 3: Some of the equipment used to rear AGM and *Trichogramma*. (**A**) Moth units showing two tiers of cribs and collection bottle. The nylon screen at the front of the unit has been lifted for this photograph. (**B**) Morrison box for rearing *Trichogramma*. (**C**) AGM oviposition unit.

Following infestation the cribs were placed 100 mm apart on steel horizontal storage racks to allow the AGM eggs to hatch, and the larvae to penetrate the grain and develop. After three weeks the cribs were transferred to the moth collection units and stored vertically to collect AGM moths as they emerged from the grain.

Temperature was maintained at 25±2°C and relative humidity at 70±10% in the AGM rearing room. Lights were used when the room was occupied by working staff (usually 6 hours maximum per day).

Adult AGM commenced emerging approximately 21 days after infestation. The moths were blown into the plastic collection bottles at the bottom of each moth unit with a vacuum cleaner and were collected daily for five weeks. Used wheat was dumped and the cribs were washed and dried in the sun.

The moths were placed into stainless steel wire mesh (8 strands/cm, 26 SWG, 0.8 mm aperture) cylinders (240 mm long, 290 mm Ø) with plastic ends. A central, threaded stainless steel rod (6 mm Ø) held the plastic ends against the wire mesh. A hole (45 mm Ø) was drilled in one end of each cylinder to allow collection bottles to be emptied via a plastic funnel into the cylinder daily, and was sealed with a cork stopper.

The oviposition cylinders were placed in an enclosed unit (1040x730x640 mm) (Figure 3c). Two 6 mm thick clear acrylic bi-fold doors (1040x600 mm) at the top of the unit allowed a maximum of six cylinders to be placed in two rows on stainless steel rods (19 mm Ø) that were connected to a geared motor that rotated the cylinders four times per minute. A moth scale extraction fan was connected to a stainless steel tube (100 mm Ø) containing inlets located 3 mm from the top of cylinders placed in the unit. The fan removed moth scales from the top of the cylinders without drawing moths or eggs (at the bottom of the cylinders) from the unit. The fan and motor were connected to a timer with a manual over ride switch, and operated automatically for two minutes every hour. The manual over ride was used for five minutes when moths were first added to a cylinder to remove excess scales.

The tumbling of the moths dislodged the eggs, which fell through the wire mesh and onto stainless steel collection trays (570x200 mm) at the bottom of the unit. A second vacuum system continually drew a slight airstream over the collection trays to withdraw any minute moth scales falling out of the cylinders. AGM eggs were collected daily and sieved through a series of stainless steel mesh screens (9 strands/cm, 13 strands/cm, and 33 strands/cm). The finest screen separated mite (*Blattisocius keegani* Fox) eggs from AGM eggs. AGM egg production fluctuated throughout the project (Figure 4).

Trichogramma and relatives

After collection, AGM eggs were held at 7±1°C to retard development. AGM eggs held for up to 28 days at this temperature could still be used to rear parasitoids. When needed, AGM eggs were weighed and sprinkled uniformly through a stainless steel wire screen (16 strands/cm) onto a white plastic sheet (370x226x2 mm) misted with distilled water. A natural

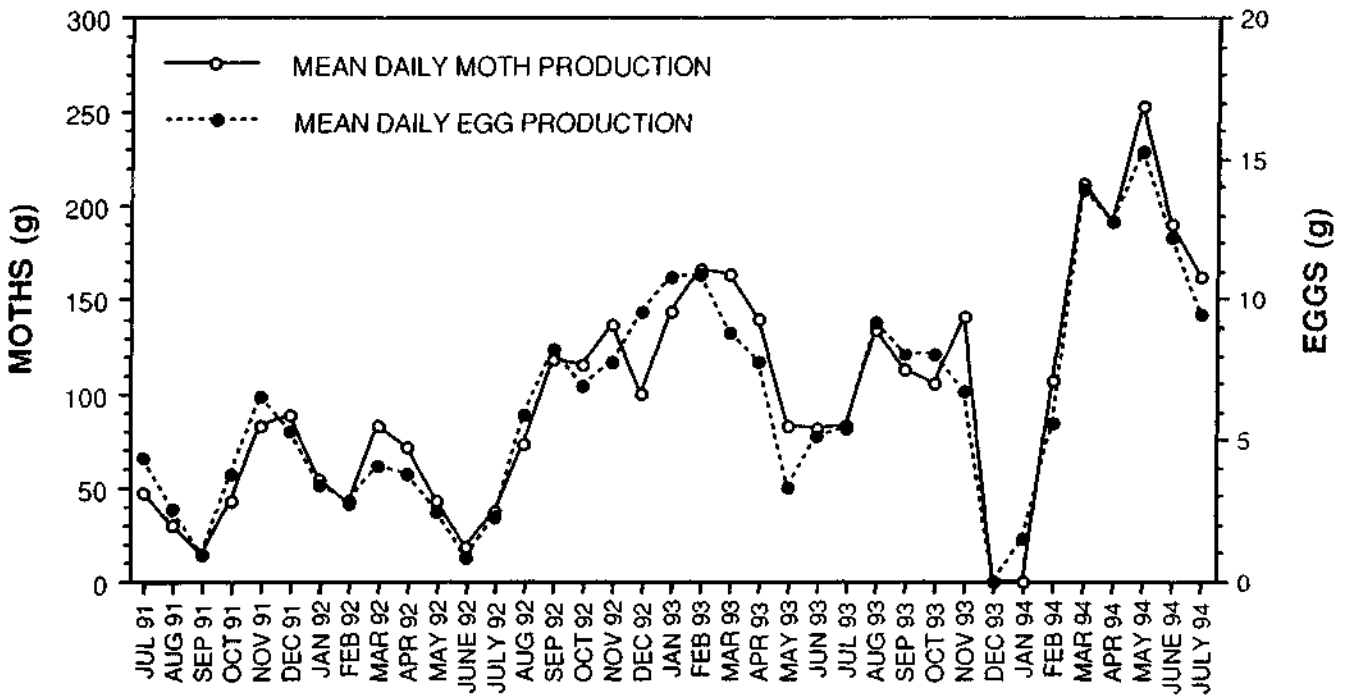

FIGURE 4: Angoumois Grain Moth (AGM), *Sitotroga cerealella*, egg production since the commencement of the project. There are approximately 50,000 eggs per gram.

adhesive on the eggs stuck them to the plastic when they contacted moisture. After drying, eggs could be easily removed with a soft paint brush held diagonally to the plastic sheet and moved downwards in short, gentle strokes.

Parasitoids were reared in a Morrison type unit (Morrison, 1985a) (Figure 3b). The unit (762x540x250 mm) contained 10 shelves. The sides were made from 10 mm thick clear acrylic sheets (750x527 mm), and the top, bottom and ends were made from 6 mm thick black acrylic. The top and bottom (750x250 mm) were glued to the sides with 'Weld On #16' acrylic glue. Nine removable black acrylic shelves (750x240x3 mm), each 50 mm apart, were fitted into routed channels (3 mm wide and 5 mm deep) in the clear acrylic sides. All black acrylic was sanded with fine sand paper to give a dull, non-reflective, lustre. Ventilation holes (25 mm Ø) were drilled in the clear acrylic sides between each shelf channel and 175 mm from each edge. The ventilation holes were covered with fine stainless steel mesh (47 strands/cm, 44 SWG, 0.125 mm aperture) to prevent parasitoid escape while allowing ventilation. Hinged, removable black acrylic doors (540x250 mm) attached by case catches were fitted to both ends of the unit. One half of each clear acrylic side was painted with flat black paint to create a unit consisting of a 'dark' end and a 'light' end. The clear acrylic on the light end was also sanded with fine sand paper to give an opaque lustre.

During operation, parasitoid pupae ('sting stock') were placed in the dark end of the unit to provide approximately one parasitoid per 5 AGM eggs. AGM eggs stuck to white plastic sheets were placed in the light end of the unit. Emerging parasitoids were positively phototactic and moved to the light end of the unit where they encountered unparasitised AGM eggs. The sting stock were removed from the dark end after 24 hours of parasitisation, and a new AGM egg sheet was inserted into the light end pushing the previous day's egg sheet to the dark end. The parasitoids then moved from the dark end to the light end and again encountered unparasitied eggs. This process could be repeated for the life of the parasitoids (usually 4-5 days).

The unit was operated in a constant temperature room (25±2°C, 70±10% R.H.) with constant artificial light provided by an 18W fluorescent light (600 mm long) supported 200 mm from each side of the light end of the unit on a steel frame.

Parasitised AGM eggs turn black after four days (unparasitised eggs are white or red, depending on their stage of development) and adult wasps emerge after another six days at 25°C. Parasitised hosts can be removed from the egg sheets with a soft paint brush, weighed, and stuck to paper cards with a water soluble glue (Aquadhere) for use in field release programs or as sting stock to increase parasitoid numbers. Parasitised host eggs weigh slightly less than unparasitised hosts (54,900 and 50,400 eggs per g respectively). Weights can be used to determine parasitoid numbers as one parasitoid emerges per parasitised AGM egg. Samples of parasitised AGM eggs should be regularly taken and the emerging parasitoids sexed and counted to determine sex ratios.

LETTUCE

Field Collections

Heliothis eggs were collected from commercial plantings of lettuce at Cambooya, near Toowoomba, to monitor heliothis infestations and natural levels of egg parasitism. A similar pattern of heliothis infestation occurred for all of the three spring seasons (1991/92-1993/94) monitored (Figure 5). Infestation peaked during the second or third week of September each year. These peaks coincided with *H. punctigera* activity reported on the Darling Downs. Of the 952 viable heliothis eggs collected only 17 (1.8%) were parasitised. Three species of parasitoids were reared from these eggs, viz. *Telenomus* sp., *Trichogramma* sp. and *Trichogrammatoidea bactrae* Nagaraja. All attempts to establish laboratory colonies of these wasps were unsuccessful due to the low numbers of parents available. Colonies of parasitoids collected from other crops were used in glasshouse trials to identify those species with promise as biocontrol agents.

Glasshouse Trials

Initial glasshouse trials with the wasps *T. bactrae* and *Trichogramma carverae* Oatman and Pinto indicated that they both attacked heliothis eggs on lettuce plants (Table 1). Although *T. bactrae* parasitised a high proportion of eggs (94%) and would therefore appear to be an ideal biocontrol agent, the results should be treated with caution because a high proportion of eggs collapsed in the trial and were excluded from analyses. A more detailed comparison of different species of native wasps was carried out.

The numbers of heliothis eggs parasitised by five species of native egg parasitoids was compared using caged lettuce plants in a glasshouse. *Trichogramma australicum* Girault, *T. carverae*, *T. funiculatum* Carver, and *T. bactrae* were collected from heliothis eggs on sorghum on the Darling Downs. *Trichogramma ivelae* Pang and Chen was collected from heliothis on peas in Victoria. Laboratory colonies of all five species were maintained on eggs of *H. armigera*.

Heliothis eggs used in the experiment were either laid onto paper towelling, cut into strips containing five eggs and stapled to the leaves (egg cards), or were laid directly onto lettuce plants by tethered moths (natural egg laid). Heliothis moths were tethered to stakes beside potted plants by gluing a cotton thread to the upper thorax. The thorax was first sanded with sand paper and the thread was attached with Norton's shearers adhesive (non-toxic to the moths).

The numbers of eggs parasitised by each species of wasp varied (Figure 6). *T. australicum* and *T. funiculatum* parasitised more eggs than the other species studied.

Effects of Insecticides

Bioassays were completed to assess the effects of insecticides and fungicides on the survival of adult egg parasitoids. Those chemicals most commonly used on lettuce were assessed at the registered rates of application.

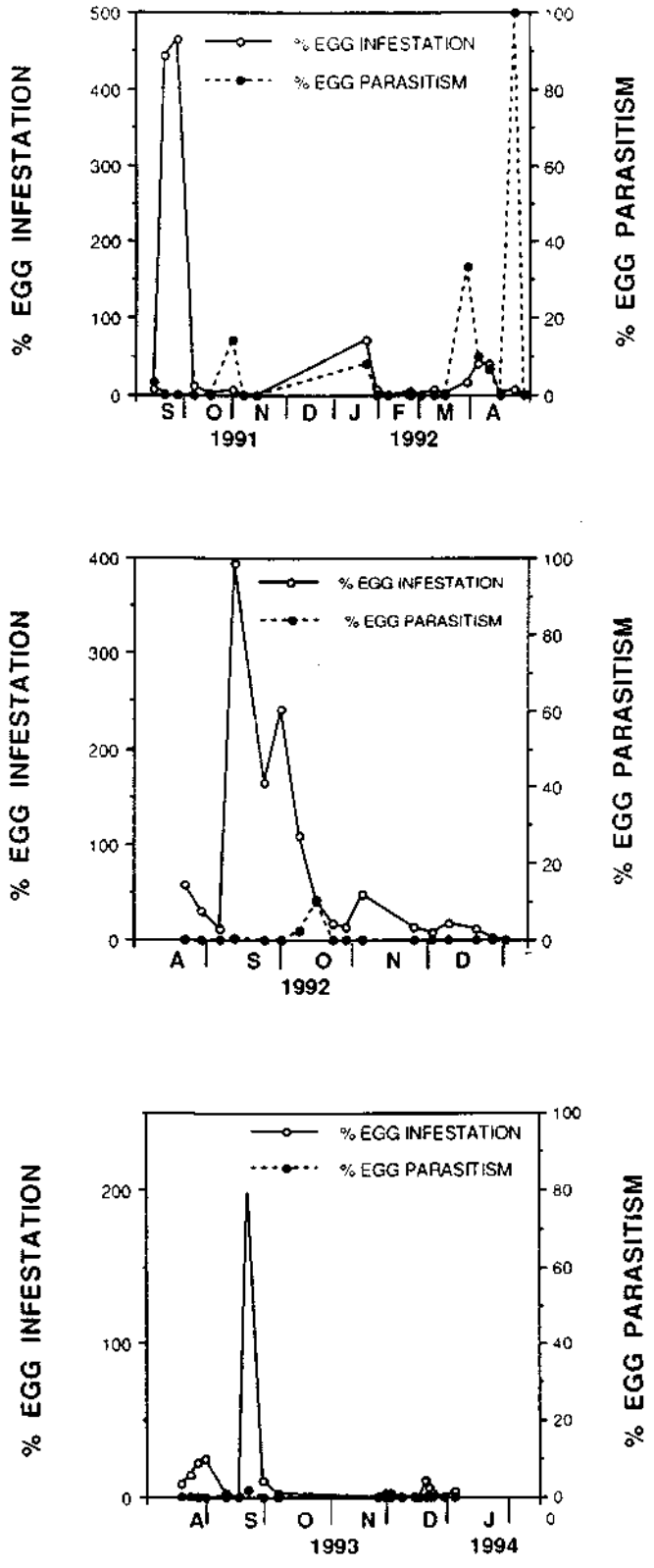

FIGURE 5: Heliothis infestation and egg parasitism on lettuce at Cambooya (S27°45' E 151°53') during three seasons (1991/92 - 1993/94).

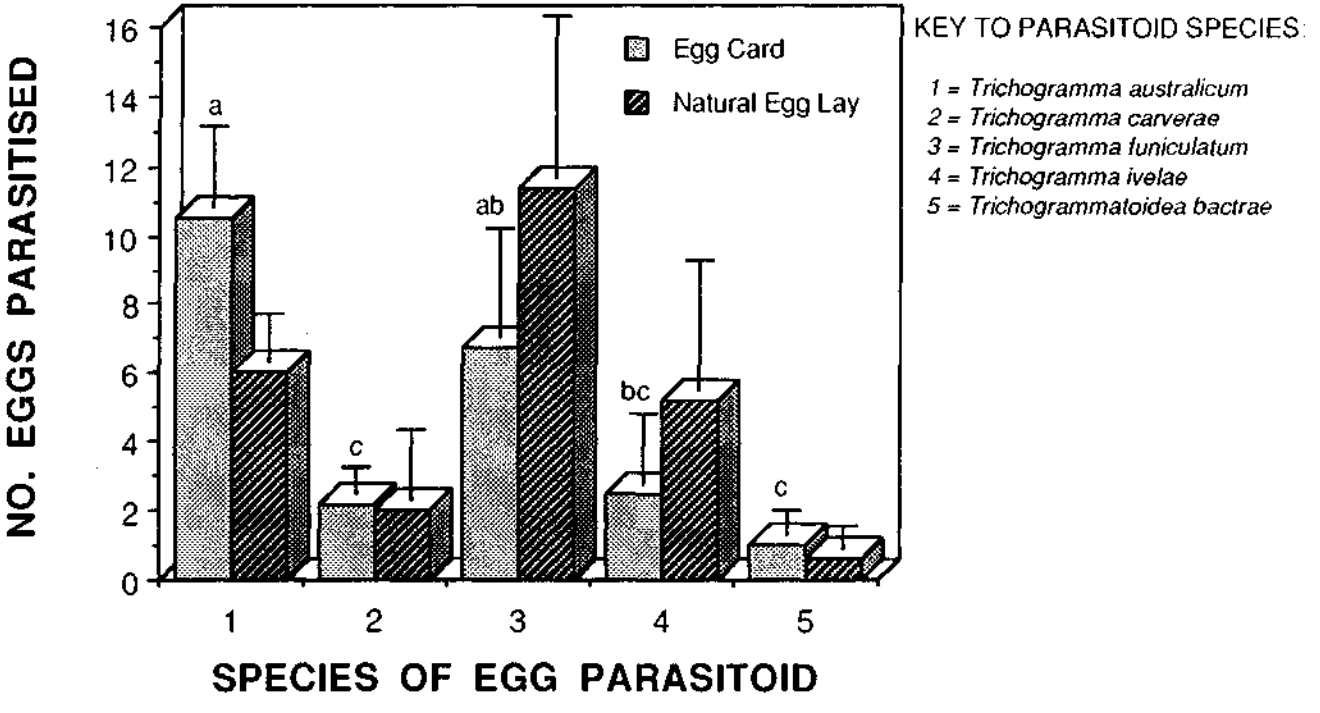

FIGURE 6: The number of heliothis (*H. armigera*) eggs parasitised by five different species of egg parasitoids on potted lettuce (var. Seagreen) plants in a glasshouse. Heliothis eggs were either laid on paper cards (Egg Cards) which were stapled to plants, or laid onto plants by tethered moths (Natural Egg Lay). All eggs were approximately 24 hours old when parasitoids were released. Cages were placed over plants and four female parasitoids were released into each cage. Parasitoids were allowed to search for heliothis eggs for 48 hours. The eggs were then collected and held to record the numbers parasitised. Values represent the mean ± standard error of six replicates. Data were analysed by analysis of variance and means were compared by Fisher's Least Significant Difference technique. Means followed by the same letter are not significantly different for $P<0.05$. There were no significant differences between species on naturally laid eggs.

TABLE 1

Results of preliminary trials for two species of egg parasitoids against heliothis on lettuce (Classic) in a glasshouse. Heliothis moths were allowed to naturally lay eggs on potted lettuce plants. The eggs were labelled by circling them with a niko pen. Newly emerged egg parasitoids were released into the glasshouse at a rate of 2.8 females/plant. The heliothis eggs were collected after 48 hours and held in a constant temperature room (25°C and 70% R.H.) to record the proportion that hatched and the proportion that were parasitised.

Species of Egg Parasitoid	*Mean No. of Eggs/Plant*	*% Eggs Hatched*	*% Eggs Parasitised**
T. bactrae	1.9	1.5	93.9
T. carverae	4.0	56.8	14.7

* N.B. These percentages represent the proportion of VIABLE eggs. A high proportion of eggs laid by moths collapsed (74% and 33% for *T. bactrae* and *T. carverae* respectively) and were excluded from analyses.

An apparatus for testing parasitoid survival in 60 bioassay chambers was constructed (Figure 7). Each chamber consisted of a glass cylinder 150 mm long and 40 mm in diameter with flamed ends. Wasp proof stainless steel gauze (47 strands/cm, 0.125 mm aperture and 0.08 mm wire diameter) was glued to one end of each chamber. Sprayed cotton leaves and parasitoids were introduced via the open end which was then sealed with a single-hole rubber stopper. A 6 mm diameter plastic tube through the rubber stopper linked each chamber to a 25 mm diameter PVC pipe that was connected to an air pump. An airflow of 38 ± 2 cm/sec was pumped through all chambers to remove any possible fumigant action of the insecticides being tested. Wasp proof gauze was also melted onto the end of the plastic tube that opened into each chamber. This ensured that parasitoids were contained within each chamber. The survival of two species of egg parasitoids, *T. bactrae* and *T. carverae*, were assessed (Figure 8).

There was no significant difference between survivorship on leaves sprayed with *Bacillus thuringiensis* (Dipel) or chlorfluazuron (Helix) when compared to a control sprayed with water. This indicates that Dipel and Helix are compatible for use with egg parasitoids. Survivorship on leaves sprayed with endosulfan, permethrin or methomyl was not high, particularly up to 11 days postspray, indicating that parasitoids are susceptible (and not compatible) with these chemicals. It should be noted that the breakdown of chemicals in glasshouses occurs slower than in the field. Survival may, therefore, be higher under field conditions. However, it is clear that Dipel

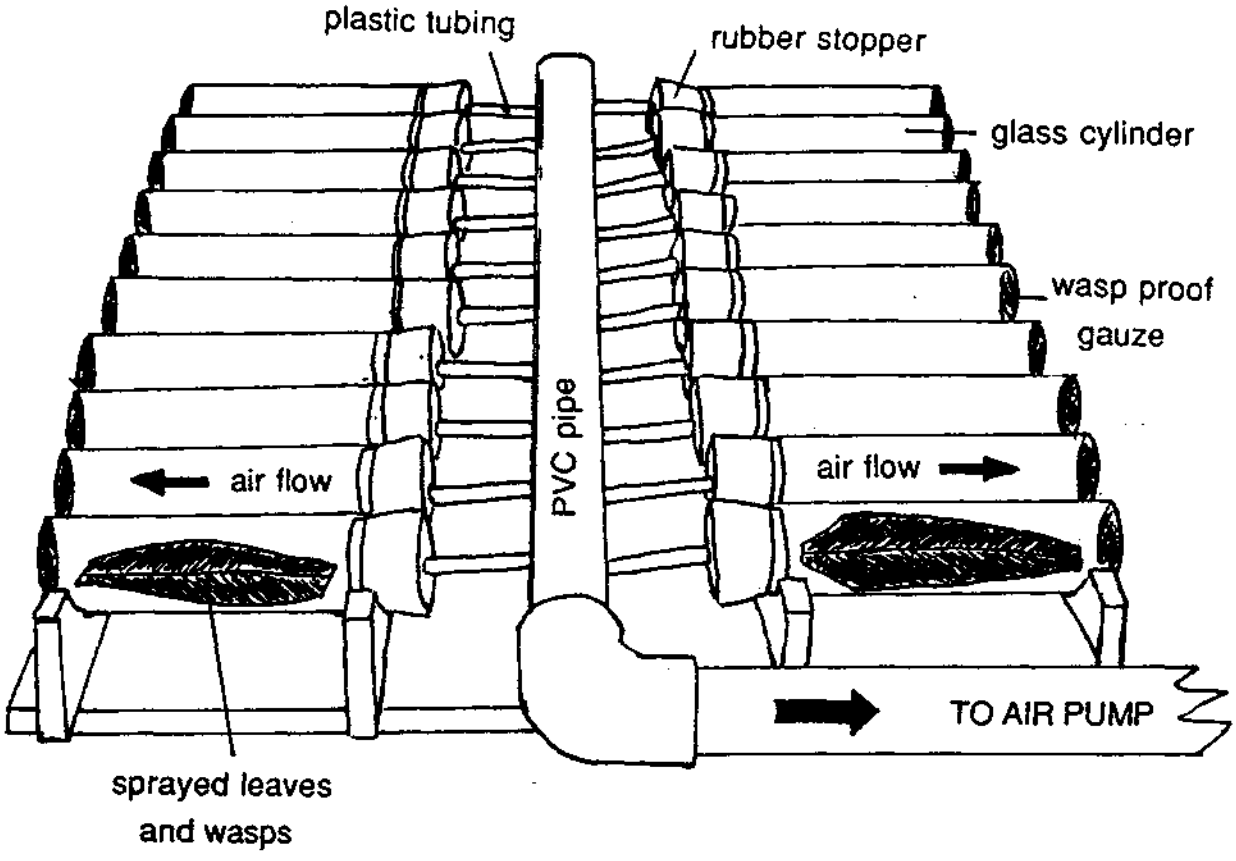

FIGURE 7: Bioassay chambers used to study wasp survival on sprayed lettuce leaves.

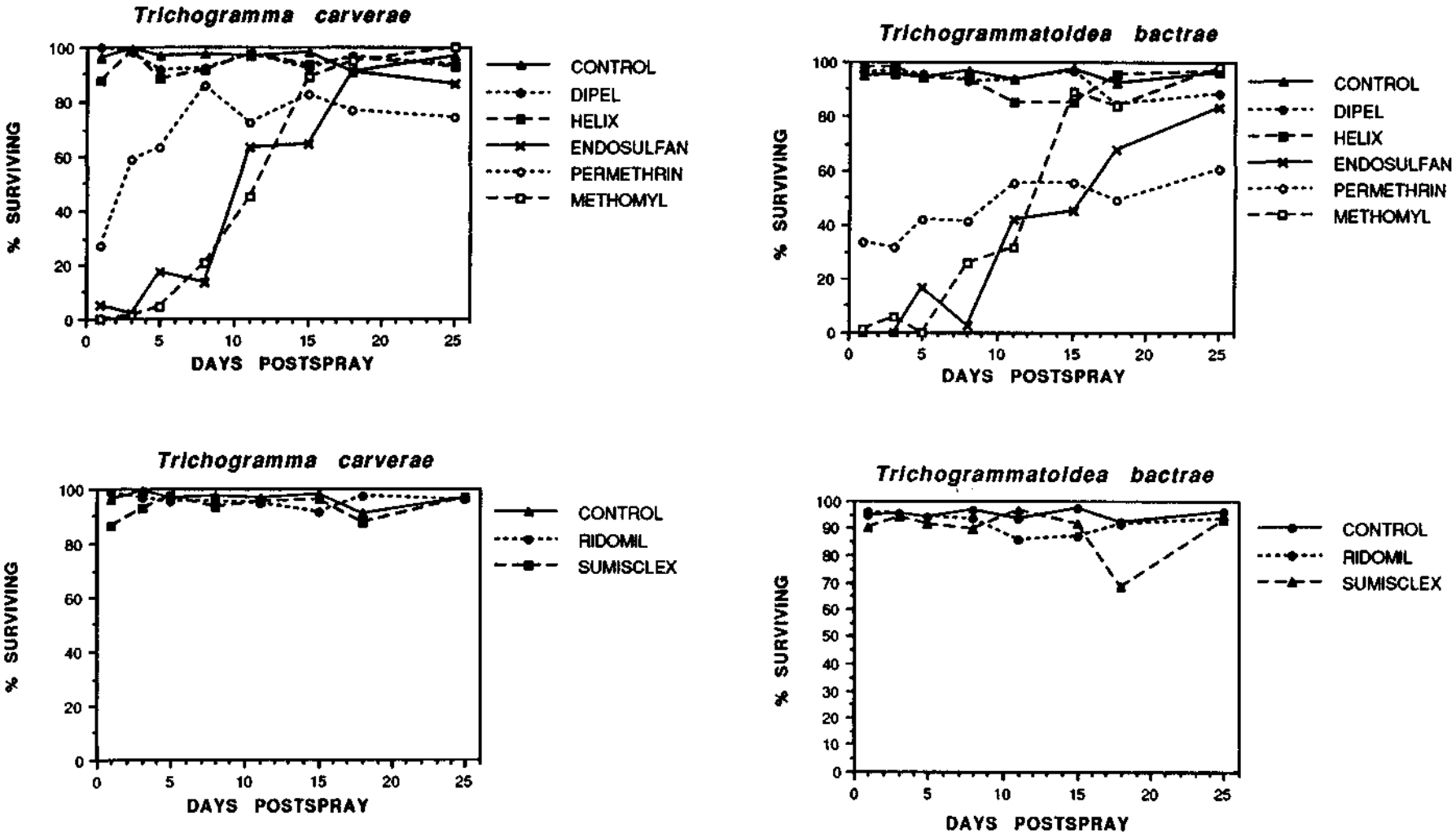

FIGURE 8: The effects of different insecticides (top) and fungicides (bottom) on the survival of two species of egg parasitoids. Insecticides were applied to potted lettuce plants in a glasshouse. Leaves were removed 1, 3, 5, 8, 11, 15, 18 and 25 days after spraying and placed in glass tubes. A constant airflow of 38 cm/sec was passed through the tubes. Egg parasitoids (10 adults of each species) were released into the tubes. The numbers of parasitoids surviving after two hours were recorded and the average % survivorship was calculated. Each treatment was replicated ten times. Rates of chemicals: *Bacillus thuringiensis* var. *kurstaki* (Dipel) - 0.5 g/L; chlorfluazuron (Helix) - 2 ml/L; endosulfan (Thiodan 350 g/L) - 2 ml/L; permethrin (Ambush EC 500 g/L) - 0.2 ml/L; methomyl (Nudrin 225) - 2 ml/L; metalaxyl+mancozeb (Ridomil MZ 250) - 2.5 g/L; procymidone (Sumisclex 250) - 2.2 ml/L; control - water.

TABLE 2

The numbers of heliothis eggs collected and levels of parasitism recorded at Bundaberg 14-15 April 1993. *T. ivelae* was obtained from commercial insectaries and released regularly at the release site. The non-release (other) sites were commercially managed, i.e. insecticides were used for heliothis control.

Site	*No. Eggs Collected*	*No. Eggs Hatched*	*Eggs Not Hatched*	*No. Eggs Parasitised*	*% Viable Eggs Parasitised **
Release	142	88	10	44	33.3
Other	590	275	289	26	8.6

* Viable Eggs = Total Eggs - Eggs not Hatched.

TABLE 3

The identity of egg parasitoids reared from heliothis eggs on tomatoes at Bundaberg 14-15 April 1993. The species of *Trichogramma* are difficult to distinguish and were classified according to colour. *T. ivelae* was released at the release site throughout the season and is a dark *Trichogramma.* Values represent the percentage of each species of wasp.

Site	Dark *Trichogramma*	*T. bactrae* *	*T. robusta* *
Release	39.6	25.6	34.8
Other	31.5	63.2	5.3

* Both are species of *Trichogrammatoidea.*

and Helix are safe for use with parasitoids. The fungicides Ridomil and Sumisclex did not adversely affect wasp survival.

Field Trial

Approximately 7,200 lettuce seedlings (Classic) were transplanted at Cambooya on November 16, 1993 for an inundative release trial. These plants were not sprayed with chemical insecticides, and were specifically planted away from other crop to avoid chemical drift. There was very little heliothis activity during Nov.-Dec. 1993 and few eggs were laid in the trial site (Figure 5). Consequently the trial had to be abandoned because there were not enough heliothis eggs in the crop to assess the impact of inundative *Trichogramma* releases on egg mortality.

TOMATOES

Field Collections

Heliothis eggs were collected from tomatoes at Bundaberg to compare the levels of parasitism and species of wasps found at a site where parasitoids had been released (John Hall's) and other sites. The egg parasitoids used at the release site had been obtained from commercial insectaries. *Trichogramma ivelae* is available from Biological Alternatives at Moree, and *Trichogramma brassicae* Voegele is available from Bio-Protection at Warwick. The identification of these wasps has lead to confusion. It now seems likely that both insectaries are rearing the same species of wasp. The species was originally described as being *T. ivelae*, but is now believed to be *T. brassicae*. *T. ivelae* is the species name that has been documented in the literature (e.g. McLaren and Rye 1981, 1983), and is probably the best name to use until a revision of the Australian fauna occurs.

The levels of egg parasitism at the release site were higher than the non-release sites (Table 2). This may have been due to the releases of *T. ivelae*, or to a larger natural population of wasps that built up in the absence of insecticides, or to a combination of both these factors. The identity of wasps at both sites (Table 3) suggests that natural populations of parasitoids were active throughout the region. More than 60% of wasps recovered from the release site were not *Trichogramma*, i.e. they were *T. bactrae* or *T. robusta* Nagaraja (both species of *Trichogrammatoidea* that are easily distinguished from *Trichogramma*). Dark *Trichogramma* were also collected from the non-release sites suggesting that there was a natural population of Dark *Trichogramma* active in the area. The identity of wasps is critical to understand the impact of parasitoid releases on heliothis, and needs to be more fully investigated. From the data presented here it seems certain that natural populations of egg parasitoids contribute to heliothis mortality in tomatoes. The importance of these populations in heliothis regulation is not fully understood. We now need to clarify the role of wasp releases as events that can suppress heliothis.

Glasshouse Trials

The numbers of heliothis eggs parasitised by six species of native egg parasitoids was compared using caged tomatoes plants in a glasshouse, viz. *Trichogramma australicum* , *T. carverae*, *T. funiculatum*, *T. ivelae*, *T. bactrae* and *Trichogrammatoidea robusta*. *T. robusta* was collected from

heliothis eggs on tomatoes at Bundaberg. Laboratory colonies of all species were maintained on eggs of *H. armigera*.

Heliothis eggs were either laid onto paper towelling that was cut into strips and stapled onto plants (egg cards), or laid directly onto plants by tethered moths (natural egg lay). Very few parasitsed eggs were recorded for all species of wasps using egg cards (Figure 9). *T. funiculatum* parasitised significantly more naturally laid eggs than most of the other species of parasitoids studied.

Field Trial

A field trial was planned at Bundaberg to assess *T. funiculatum* as a biocontrol agent on tomatoes. Unfortunately the host insect used to mass rear wasps at the DPI was invaded by a larval parasitoid (*Pteromalus cerealellae*) (Ashmead)) and *Trichogramma* production closed down from November 1993 to April 1994. Another species, *T. ivelae*, was therefore purchased from Biological Alternatives for use in the Bundaberg trial. A single release of *T. funiculatum* was made on April 22, 1994, but all other releases (a total of eight) were of *T. ivelae*. *T.ivelae* was also the species that was used in other trials in the district.

A 0.1 ha planting of tomatoes at DPI Bundaberg Research Station was used for the field trial. Wasps were released at a rate equivalent to 500,000/ha (approx. 300,000 females/ha). Egg parasitoids were mass reared on eggs of the grain moth *Sitotroga cerealella*, and were released as pupae sprinkled directly onto leaves moistened with water from a hand-held atomiser. Parasitoids were released 15 m apart in all but the outside rows, with releases commencing 5 m or 10m into the crop in alternate rows. The control site (approximately 500 m from the release site) was sprayed with *Bacillus thuringiensis* only.

Heliothis pressure was high throughout the trial (Figure 10), peaking at 81.5 eggs per 30 leaves. Wasp releases failed to manage this infestation and no fruit were harvested due to the extremely high level of damage. The levels of egg parasitism reached high levels (up to 87.7% in the release site), but only towards the end of the season (Figure 11). High levels of egg parasitism were also recorded in the control site (up to 90.1%) (Figure 11) indicating that natural populations of wasps were active. The released wasp, *T. ivelae*, is a dark *Trichogramma*. Dark *Trichogramma* were dominant in both the release and control sites suggesting that there may have been a natural population of wasps that resembled the released wasps. The impact of the wasp releases is therefore unclear.

These data suggest that natural populations of wasps may have been responsible for parasitism in the release site because the majority of the nine wasp releases failed to increase levels of parasitism markedly. The levels of egg parasitism recorded in the release site may have been the result of a build up of natural populations of wasps in the absence of insecticides instead of inundative releases. Further research is needed to clarify the role of natural and released wasp populations.

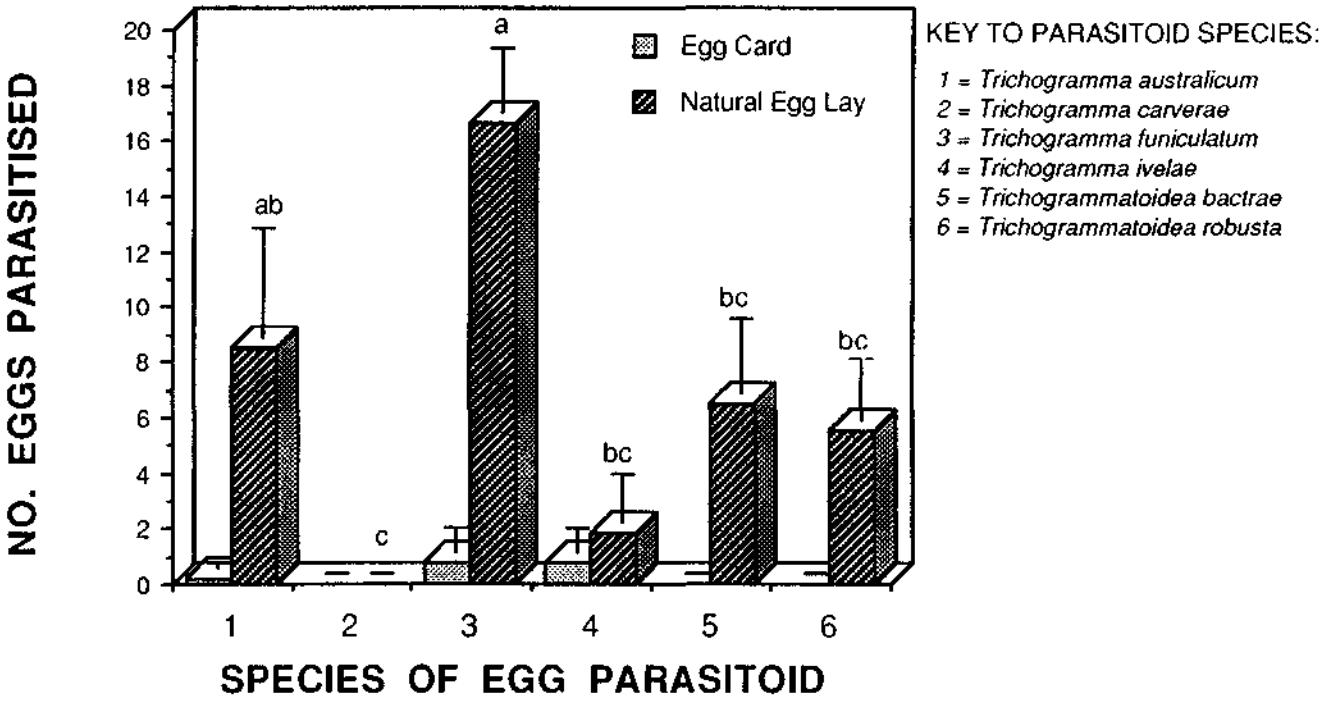

FIGURE 9: The number of heliothis (*H. armigera*) eggs parasitised by five different species of egg parasitoids on potted tomatoes (var. Floradade) plants in a glasshouse. Heliothis eggs were either laid on paper cards (Egg Cards) which were stapled to plants, or laid onto plants by tethered moths (Natural Egg Lay). All eggs were approximately 24 hours old when parasitoids were released. Cages were placed over plants and four female parasitoids were released into each cage. Parasitoids were allowed to search for heliothis eggs for 48 hours. The eggs were then collected and held to record the numbers parasitised. Values represent the mean ± standard error of six replicates. Data were analysed by analysis of variance and means were compared by Fisher's Least Significant Difference technique. Means followed by the same letter are not significantly different for $P<0.05$. There were no significant differences between species on egg cards.

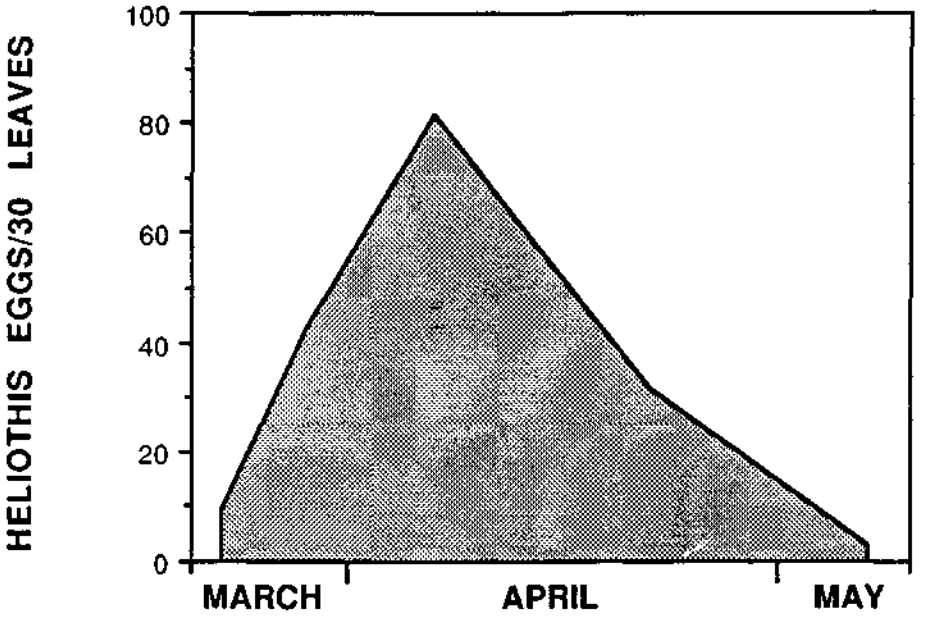

FIGURE 10: Heliothis egg infestation on tomatoes at Bundaberg 1994.

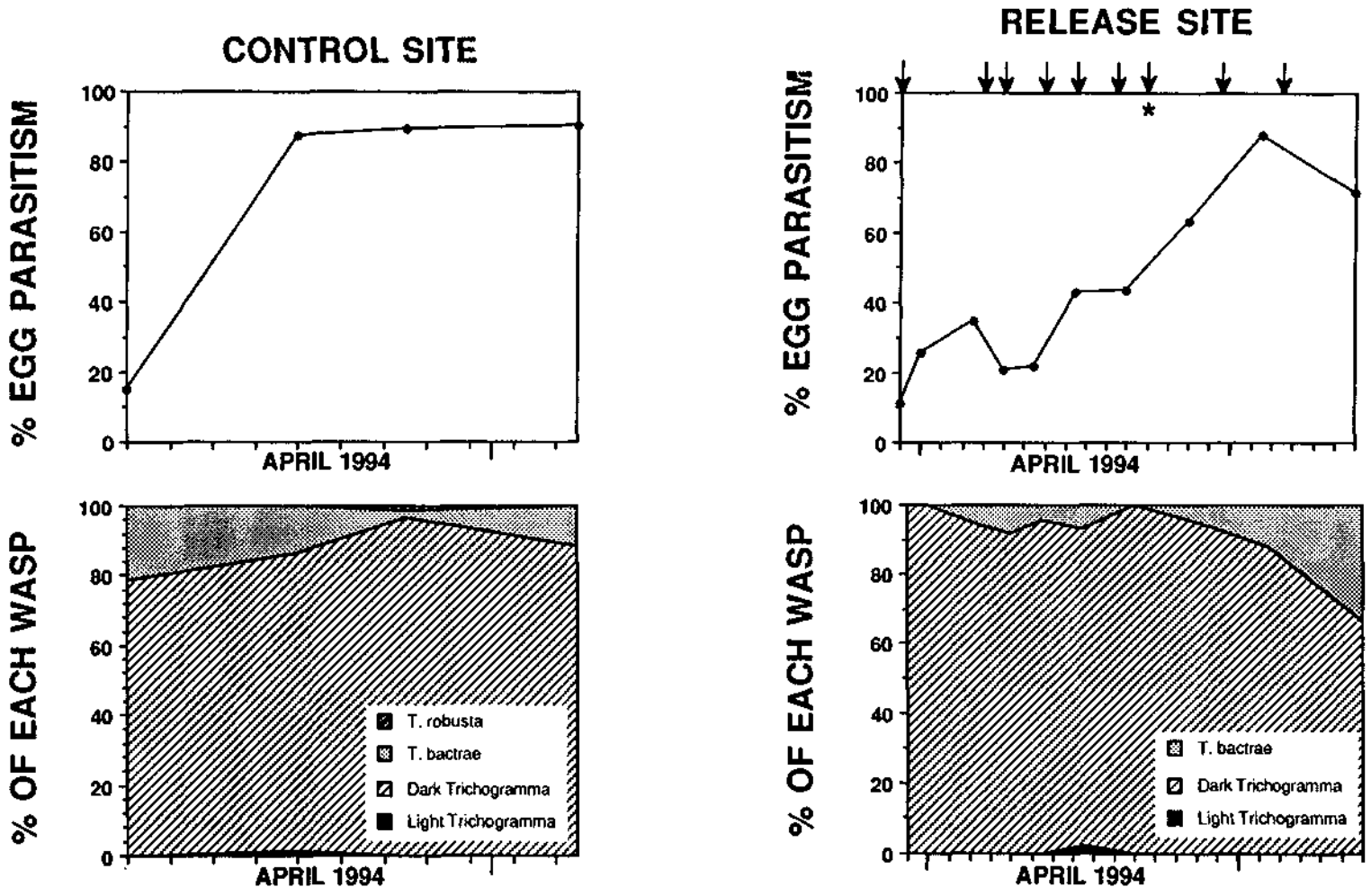

FIGURE 11: The levels of heliothis egg parasitism (top) and proportion of each species of wasp found (bottom) on tomatoes at Bundaberg. Arrows indicate when *T. ivelae* and *T. funiculatum* (*) were released at a rate equivalent to 500,000/ha.

SWEET CORN

Field Trials

Egg parasitoid releases were carried out against heliothis on sweet corn (Florida) at Grasstrees on the Darling Downs. Two bays of corn (approx. 0.64 ha) were treated with parasitoids. One of these bays was also treated with *Bacillus thuringiensis* (Dipel). An adjacent two bays were commercially managed for heliothis i.e. sprayed with thiodicarb (1.5L) every two days.

Egg parasitoids were reared on the eggs of the Angoumois grain moth (*Sitotroga cerealella*) in an experimental insectary at the DPI in Toowoomba. Parasitised eggs were weighed (to calculate egg numbers) and stuck to paper with an aquadhere/water mixture. The egg cards were cut into strips and stapled to the undersurface of corn leaves in the field. Eggs cards were placed in every third row of corn at 20 m intervals.

Two species of egg parasitoids were released: *Trichogrammatoidea bactrae* (from the QDPI Toowoomba) was released on 27 February 1993, and *Trichogramma ivelae* (from Biological Alternatives at Moree) was released on 2 March 1993. The parasitoids were released as pupae, emerging from the egg cards in the field the day that they were attached to the corn leaves.

The experiment was repeated in maize (Pioneer C83) at Dalby on the Darling Downs using a single release of *T. bactrae*. Red silks were sampled. The release site was a downwind 0.25 ha fraction of an unsprayed 40 ha planting of irrigated maize. Heliothis eggs were collected from the release site and from an upwind section of the remainder of the crop.

The number of eggs rarely exceeded 1.0 / silk at Grasstrees throughout the study (Figure 12A). The level of egg parasitism reached a maximum of 23.1% in the bay where parasitoids only were released (Figure 12 B). Egg parasitism in the commercial bays were zero. The egg hatch was high in all bays (Figure 12C), including the commercial bay where a minimum of 53.1% of eggs hatched. *T. bactrae* was the most common parasitoid collected after the releases, accounting for 71.4% of parasitised eggs collected (Figure 13).

The levels of egg parasitism achieved following releases at Grasstrees were poor, and the egg hatch was high throughout (including the commercially managed bays) even though the egg infestation was not exceedingly high. There were very few *T. ivelae* parasitised eggs recovered.

The data suggest that the two-day spray regime in the commercially managed bays may have had a serious effect on parasitoid survival in the non-sprayed bays. Parasitism was highest in the bay that was most distant from the spraying (the parasites only bay in Fig. 12B), and lower in the bay adjacent to the sprayed bays (Dipel was also applied in this bay). This was probably due to spray drift from the ground rig applications in the commercially managed bays.

The two-day spray regime also had a poor impact on egg hatch, with 53-64% of eggs hatching in the commercially managed bays. This suggests

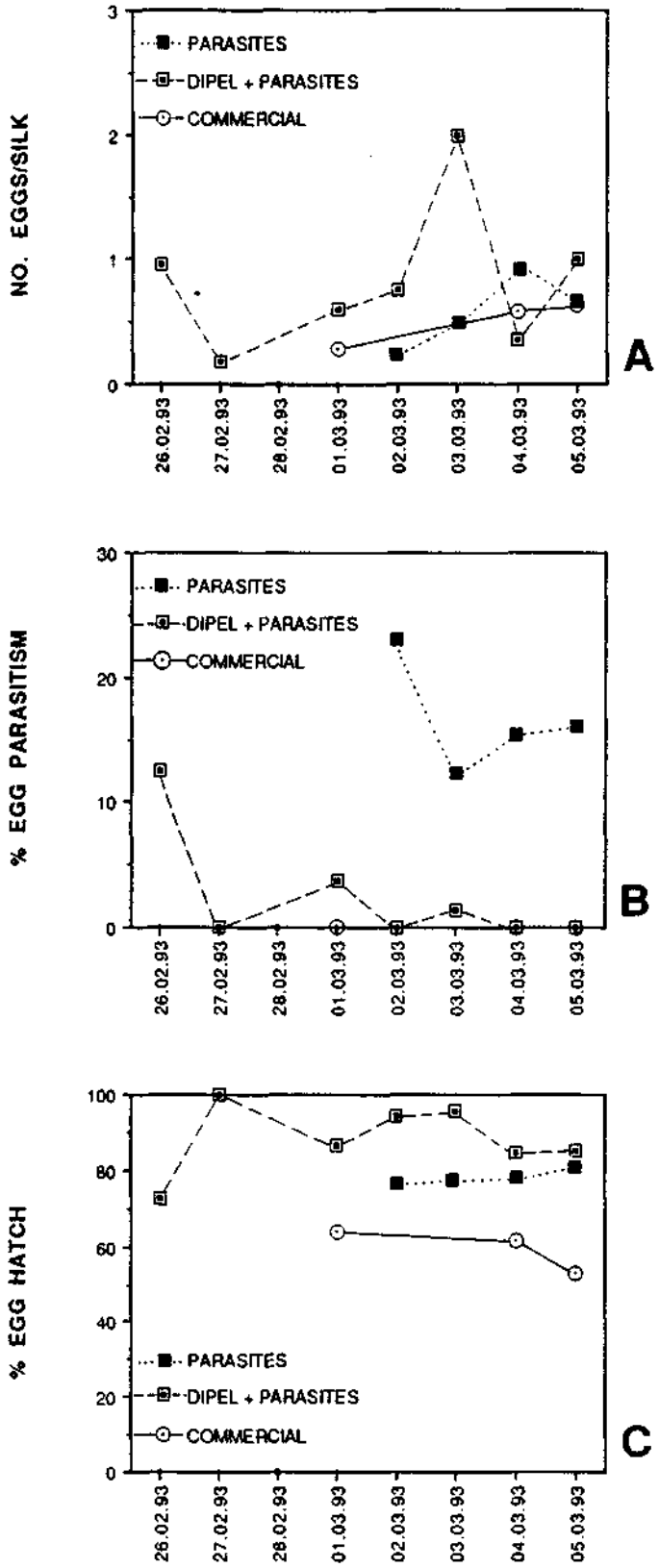

FIGURE 12: The variation of heliothis egg infestation, egg parasitism and egg hatch for sweet corn (Florida) at Grasstrees on the Darling Downs (S27°53' E151°25'). Egg parasitoids were released on 27 Feb. 1993 (*T.bactrae*: 500,000 females per hectare) and 2 Mar. 1993 (*T. ivelae:* 700,000 females per hectare). Thiodicarb (1.5 L) was applied by ground rig to the commercial site on 26 Feb., 28 Feb., 2 Mar. and 4 Mar..

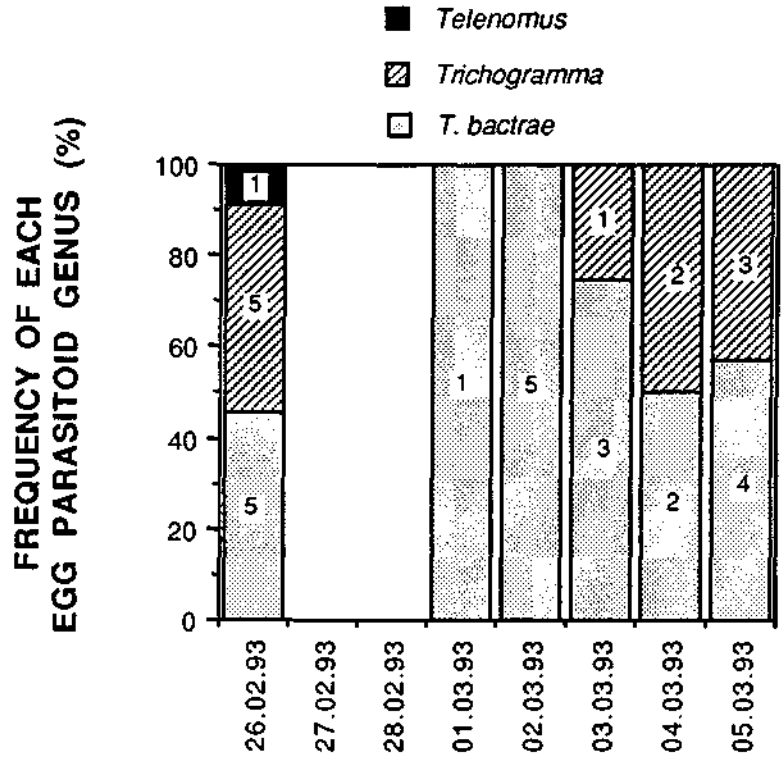

FIGURE 13: The variation in the frequency of egg parasitoid genera collected from unsprayed sweet corn at Grasstrees. Numerals within the graph bars are the actual numbers of parasitised eggs collected containing each genus of parasitoid. *T. bactrae* was the only member of the genus *Trichogrammatoidea* collected; it was not possible to accurately identify the species of *Trichogramma* and *Telenomus*. *T. bactrae* was released on 27 February and *Trichogramma* nr. *ivelae* was released on 2 March.

that an alternative method of egg management is necessary, and that egg parasitoids should be investigated more closely.

The level of egg infestation in the Dalby site was higher than that at Grasstrees (Figure 14A). The level of egg parasitism rose to 50.8% three days after the release of the parasitoids, and was negligible in the unsprayed (non-release) site (Figure 14B). Egg hatch was again high throughout the trial, although it fell from 94.7% in the unsprayed site to 41.0% in the release site three days after the release of parasitoids (Figure 14C). The level of egg parasitism and hatch were similar in both sites 10 days after the release of parasitoids (Figures 14B, C).

The results from the Dalby maize trial were encouraging. Parasitism up to 51% was recorded for egg infestations that were 2-4 times higher than those at Grasstrees. The parasitoids used in this trial (*T. bactrae* from the DPI) had been reared on *Sitotroga* for 42 continual generations and had no recent exposure to heliothis eggs. It also appeared that the release contributed to establishing parasitoids in the crop as parasitism rose from 6% on the release date to 24% 10 days after the release in a non-release site (Figure 14B).

Commercial Trial

Two releases of *T. ivelae* (purchased from Biological Alternatives, Moree) were made in a 4.5 ha of sweet corn (H5) at Mulgowie in the Lockyer Valley. Parasitoids were reared on *Sitotroga cerealella* eggs, and were released from a hopper mounted on a helicopter (the release system was developed by Biological Alternatives). Parasitoids were released at a rate of 250,000 wasps/ha. One release was made at the vegetative stage, and the other was made four weeks later at the start of the silking stage. *Bacillus thuringiensis* - molasses sprays were applied every two days at the silking stage (a total of ten sprays).

Heliothis egg densities were high during the vegetative stage, peaking at 8.1 eggs/plant (Figure 15A). The levels of egg parasitism were extremely high, ranging from 79.5 to 96.7% (Figure 15B). The level of heliothis control was good, with 85.5% of cobs being undamaged and containing no larvae. 8.3% of cobs contained larvae in the silks only and were marketable (resulting in a total of 93.3% cobs marketable). Only 6.7% of cobs contained larvae that had damaged the cob, and this damage was restricted to the top centimetre of cob.

This result is very encouraging, demonstrating that wasp egg parasitoids can manage heliothis successfully. However the source of the wasps responsible for the parasitism remains unclear. The likely source/s of the wasps were:

a) The result of the inundative releases.
b) The progeny of inundative releases previously made in the region.
c) Natural populations of wasps that resembled *T. ivelae*.
d) A combination of any of the above.

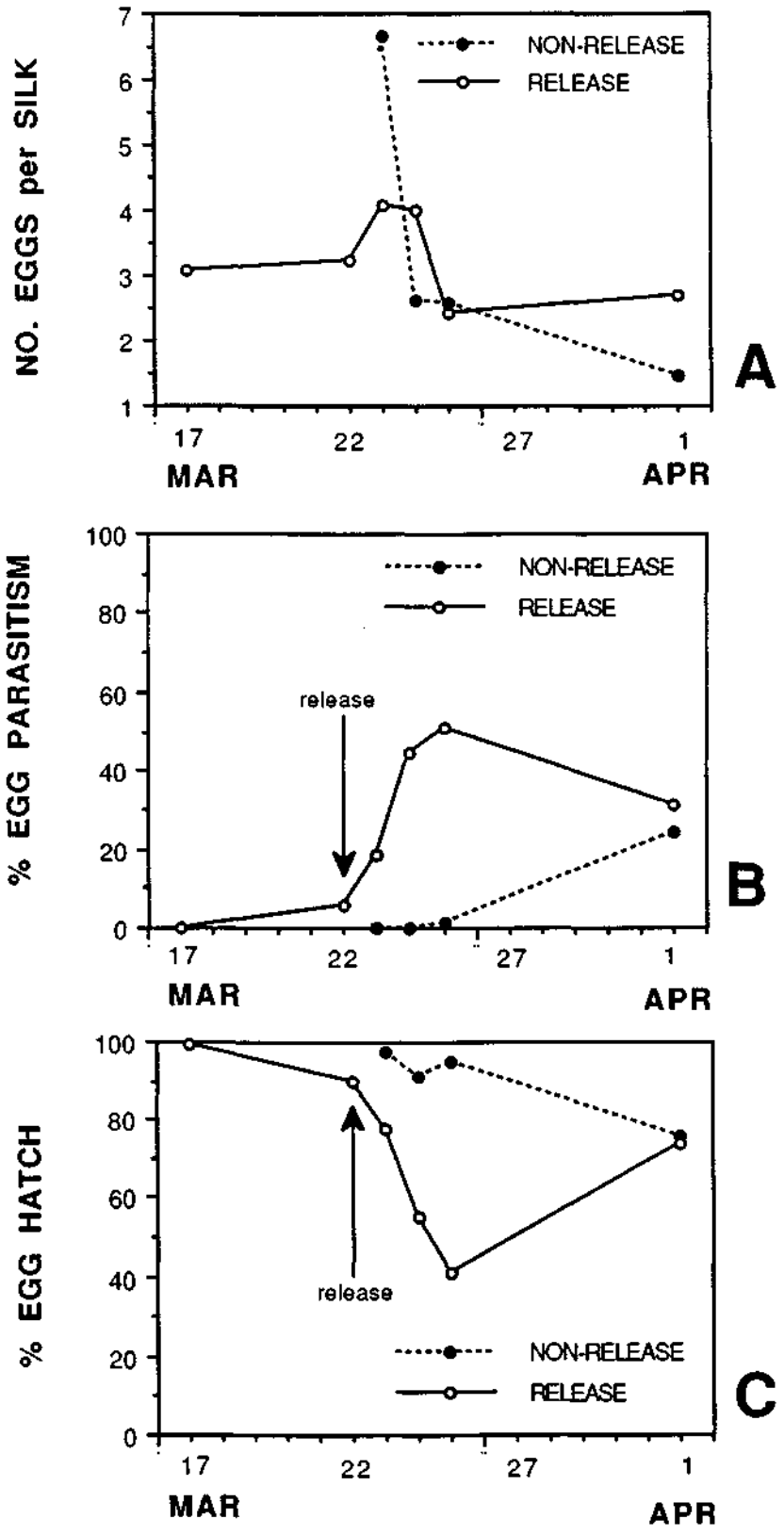

FIGURE 14: Heliothis egg infestation, egg parasitism and egg hatch for maize (Pioneer C83) at Dalby on the Darling Downs (S27°11' E151°17'). Egg parasitoids were released on 22 Mar. 1993 (*T.bactrae*: 420,000 females).

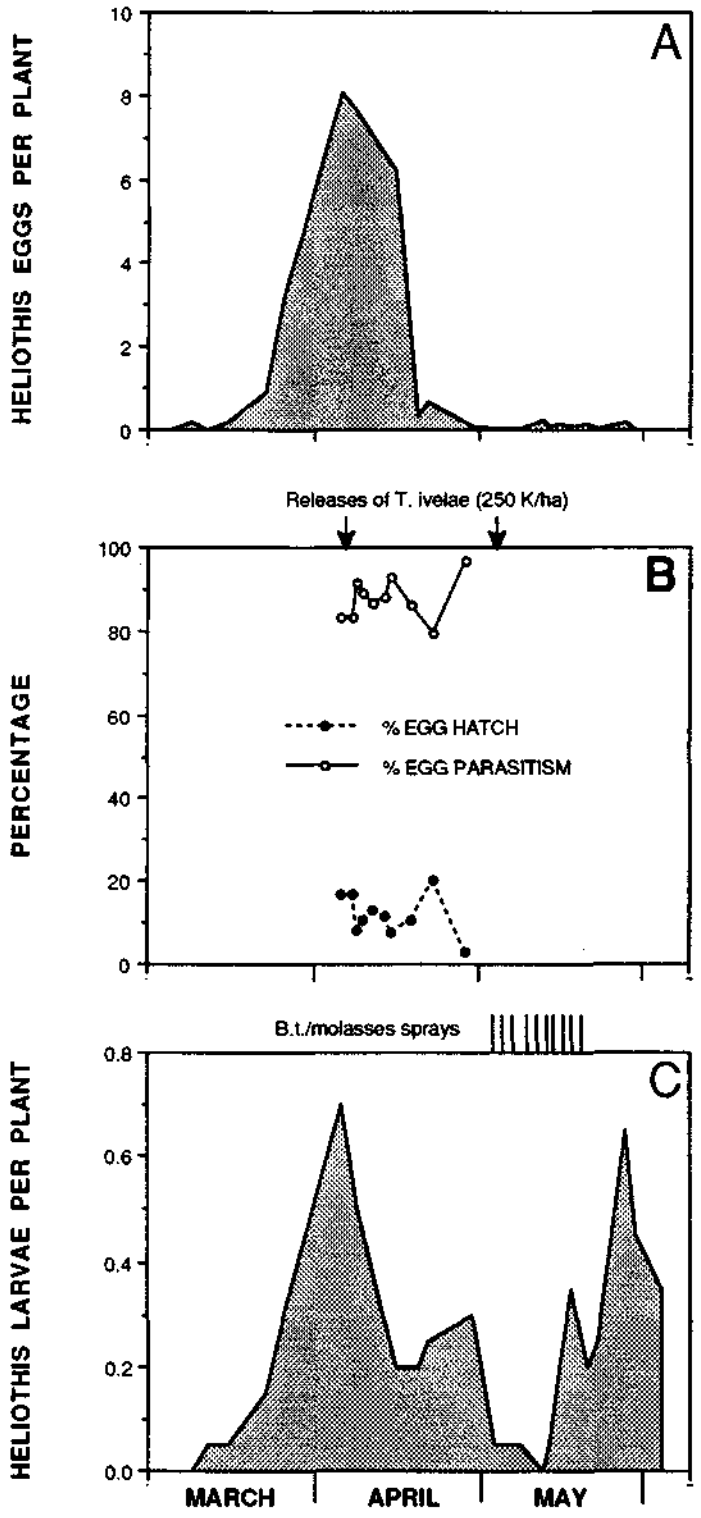

FIGURE 15: Heliothis egg infestation, egg parasitism, egg hatch and larval infestation for sweet corn (H5) at Mulgowie in the Lockyer Valley. *T. ivelae* were released on 6 April and 3 May, 1994 (250,000/ha).

As there were only two inundative releases in the trial it seems unlikely that these alone were responsible for the high levels of parasitism recorded. The progeny of previous inundative releases in the region may have contributed to some of the parasitism, but the extent of this contribution is unknown. There is some evidence to suggest that natural populations of wasps were active in the region at the time of the trial. Heliothis eggs were collected from sweet corn where no releases were made (50 km from the trial site) and 68% of these were parasitised. The majority of these wasps resembled *T. ivelae*. This suggests that natural populations of *Trichogramma* were very active, and their impact in the trial site may have been encouraged by not spraying the crop with chemical insecticides. Further research is necessary to understand the potential of natural and released *Trichogramma* as biological control agents.

DISCUSSION

Natural populations of native egg parasitoids were difficult to find in commercial plantings of lettuce on the Darling Downs (Figure 5), presumably because of the regular application of chemicals for pest management. Bioassays of residual insecticides on lettuce leaves found that the chemicals commonly used in lettuce were toxic to *Trichogramma carverae* and *Trichogrammatoidea bactrae* (Figure 8).

High natural levels of heliothis egg parasitism were found in unsprayed tomatoes at Bundaberg (Figure 11, control site). There was also indirect evidence suggesting that natural populations of egg parasitoids were active in sweet corn at Mulgowie. It seemed that natural populations of egg parasitoids had their greatest impact on heliothis towards the end of the summer growing season, particularly in crops that were not sprayed regularly with chemical insecticides. Further research is necessary to better understand the value of these natural wasp populations in heliothis management.

Inundative releases of *Trichogramma ivelae* in tomatoes and sweet corn had little impact on heliothis egg mortality. *T. ivelae* did not appear to be the most suitable species for use as a biological control of heliothis. *Trichogramma funiculatum* was found to parasitise more heliothis eggs than other species on lettuce and tomatoes in glasshouse trials (Figures 6 and 9), supporting this assumption.

The most encouraging field results were found on maize and sweet corn. A single inundative release of *Trichogrammatoidea bactrae* in maize at Dalby increased heliothis egg parasitism markedly within three days (Figure 14). Egg parasitoids and applications of *Bacillus thuringiensis* successfully managed heliothis in a late planting of sweet corn at Mulgowie (Figure 15). There was, however, some evidence to suggest that natural populations of wasps were responsible for the high levels of heliothis egg parasitism recorded. For *Trichogramma* to be accepted in pest management, future research needs to demonstrate that inundative releases of egg parasitoids can be used to manage heliothis in a similar manner. These findings do, however, indicate that egg parasitoids have potential as biological control agents of heliothis.

Further research is needed to demonstrate this potential, to promote their acceptance as a useable form of pest management, and to encourage the establishment of more *Trichogramma* production facilities in Australia.

CONCLUSIONS and RECOMMENDATIONS

The data indicate that further research is needed before an assessment of egg parasitoids for heliothis management can be made. It seems certain that native egg parasitoids have potential at managing heliothis, but their value as inundative biological control agents remains unclear.

There is evidence to suggest that *Trichogramma* have value in heliothis IPM programs, particularly when used with selective insecticides such as *Bacillus thuringiensis*. Additional research is necessary to document this potential, and to develop management guidelines.

Future research involving egg parasitoids should be directed towards the following areas:

* Additional field research evaluating different species of egg parasitoids as inundative biocontrol agents of heliothis.

* The evaluation of egg parasitoids as biological control agents in an integrated pest management program where other tactics are jointly evaluated for heliothis management.

* Knowledge of the biology and ecology of natural and released populations of egg parasitoids in the field to better understand their value as biological control agents.

* Detailed taxonomic studies of the Australian egg parasitoid fauna so that released and natural wasp populations can be distinguished reliably.

The research, development and adoption of non-chemical forms of pest management is necessary in vegetables to avoid the serious problems that will arise when chemical insecticides become unavailable or unsuitable.

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APPENDIX A

BUDGET

This research was jointly funded by the Horticultural Research and Development Corporation (HRDC) and the Queensland Fruit and Vegetable Growers Association (QFVG).

Fin. Year	HRDC	QFVG	Total
1991/92	$10,600	$10,600	$21,200
1992/93	$11,400	$11,400	$22,800
1993/94	$11,450	$11,450	$22,950
TOTAL	$33,475	$33,475	$66,950

APPENDIX B

COMMUNICATION OF RESULTS

Scholz, B.C.G. and Webster, D.E. (1993). Sprays and Egg Parasitoid Survival. *24th Australian Entomological Society Conference.* Matson Plaza Hotel, Cairns, 3-8 July, 1993. Poster.

Scholz, B.C.G. and Webster, D.E. (1994). An evaluation of an egg parasitoid for heliothis control in maize. *Proceedings of the Second Australian Maize Conference.* University of Queensland, Gatton College, February 6th-9th, 1994, pp. 73-76.

APPENDIX C

Commercial producers of *Trichogramma* in Australia.

PRODUCER	CONTACT	ADDRESS/PHONE/FAX
Bio-Protection	Richard Llewellyn	P.O. Box 35 Warwick Qld. 4370 ph: 076 661592 fax: 076 661639
Biological Alternatives	Glenn Bowman	P.O. Box 236 Moree NSW 2400 ph.: 067 549441 fax: 067 549470
National Insect Laboratories	Ross Munro or Peter Birch	Moree NSW 2400 ph.: 067 511337 fax: 067 524792