

**Production attributes of *Trichogramma* reared on Eri silkworm eggs vis-à-vis *Corcyra* eggs and economics of rearing system**

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**Attended International Training Course on the “Theory and Technology of Plant Protection for Sustainable Agriculture in 21st Century” from 2<sup>nd</sup> to 21<sup>st</sup> September, 2004 in the Shenyang Agricultural University, Shenyang, China**

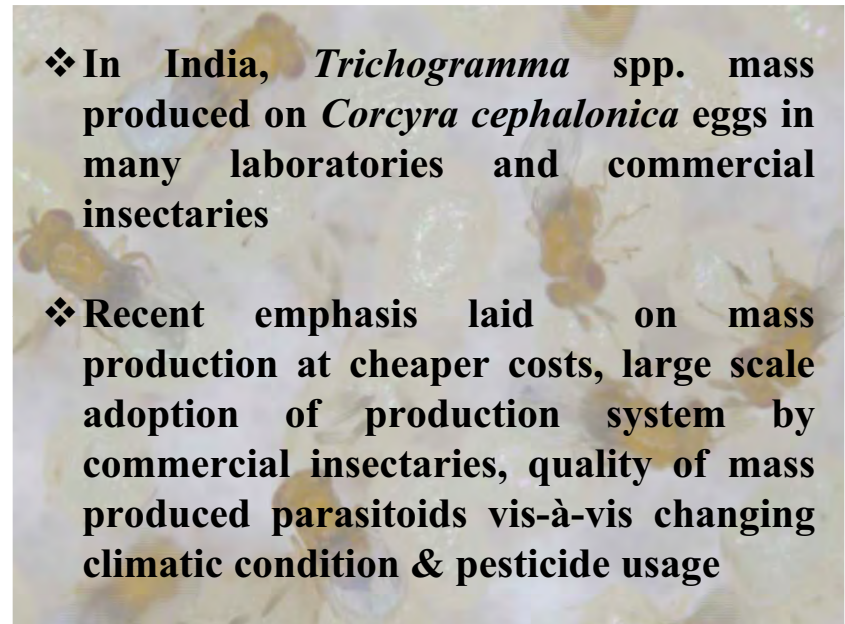
**As a consequent of the International training is the study of production attributes of *Trichogramma* reared on Eri silkworm eggs vis-à-vis *Corcyra* eggs and economics of rearing system**

❖ **Egg parasitoid – *Trichogramma* spp. mass produced on eggs of *Corcyra cephalonica*, *Sitotroga cerealella*, *Anagasta kuehniella* in major laboratories of the world**

❖ **Oak silk worm – *Antheraea pernyi* and Eri silkworm – *Samia cynthia ricini* eggs used for production of seven species of widely released *Trichogramma* spp. in China**

❖ **In India, *Trichogramma* spp. mass produced on *Corcyra cephalonica* eggs in many laboratories and commercial insectaries**

❖ **Recent emphasis laid on mass production at cheaper costs, large scale adoption of production system by commercial insectaries, quality of mass produced parasitoids vis-à-vis changing climatic condition & pesticide usage**



### Use of *Trichogramma*

- *Trichogramma chilonis* a most dominant native egg parasitoid of south-east Asia and Pacific region & most widely released in different ecosystems in India
- *T. chilonis* released in 17 crops including cereals, vegetables, commercial crops and fruit trees
- *T. japonicum* released in sugarcane, paddy & brinjal
- *T. pretiosum* (Thelytokous & Arrhenotokous) released in tomato
- *T. embryophagum* field released in apple orchard for control of codling moth

### Advantages of Eri silkworm

- ❖ Eri silkworm is an alternative host
- ❖ Feeds on castor (*Ricinus communis* Linn.)
- ❖ Requirement of minimum infrastructure man power & cost
- ❖ Mostly disease free, production on large scale & supply of parasitoids with better biotic attributes

o Eri silkworm culture - Department of Sericulture, Bangalore University, Bangalore, Karnataka, India

o *Trichogramma chilonis* indigenously collected, taxonomically confirmed with voucher specimens available in the insect collections of National Bureau of Agriculturally Important Insects, Bangalore, Karnataka, India

o Host and parasitoid cultures reared continuously in the laboratory

### Parameters studied

Acceptability  
Adaptability  
Evaluation  
Mass production  
Cost Economics



## Initial testing

- ✓ Fresh eggs exposed to *T. chilonis*
- ✓ 50-75% hatching
- ✓ Few eggs blackened
- ✓ Very few eggs with incomplete cutting of chorion
- ✓ Eleven days developmental period
- ✓ Only ten adults emerged & 70% were females
- ✓ Adult longevity 8 days
- ✓ On *Corcyra* eggs - 95.14% parasitism

## Sterilization of eggs

- i) UV treatment - 30 min, 1h, 1.30h, 2h, 2.30h - 58-78% hatching
- ii) Influence of distance (30cm from UV source) - 30 min, 1h, 1.30 h - 62-90% hatching
- iii) Storage at 4°C for 5 different time durations – 1, 6, 12, 18, 24 h - 6h storage resulted in no hatching

## Treatment of eggs and its influence on parasitism

Formalin (2%)	Hatching 38-98%	38.8 % parasitism with very less adult emergence
KOH (1%)		Completely dissolved chorion
NaHCl (0.05%)		No parasitism
Water		No parasitism

KOH (0.5%)	1, 5, 10, 15, 20, 25, and 30 min	No hatching & parasitism
KOH (0.1%)		18-44 % hatching & 58.4% & 10.4% parasitism in 1 and 30 minute treatments respectively
<b>KOH (0.1%)</b>	<b>% Hatching</b>	<b>% Parasitism</b>
1female : 5 eggs	47.7	52.3
1female : 2 eggs	31.2	58.4

## Effect of storage methods and duration on parasitism

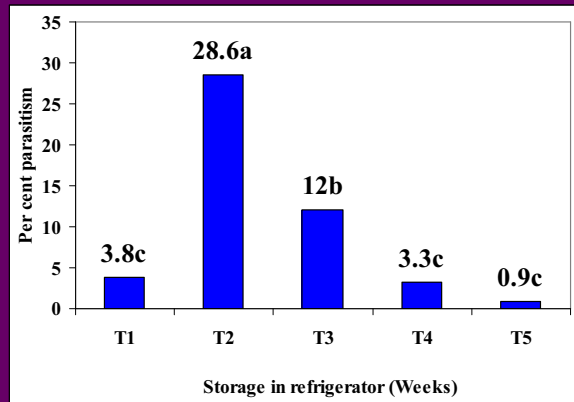
### i) Vacuum packing:

Vacuum 20, 40, 60, 80, and 100%  
N<sub>2</sub> 20, 40, 60, 80 and 100%  
Vacuum & N<sub>2</sub> 60, 80 and 100%;  
N<sub>2</sub> & vacuum 60, 80 and 100%  
Storage at 3 different temperatures  
Laboratory (27 ± 0.5°C)  
BOD (16°C)  
Refrigerator (9°C)  
Duration 5, 10, 15 and 20 days

### ii) Refrigerator (9°C)

Duration (weeks) – 1, 2, 3, 4 and 5

- Vacuum packing and storage  
Lab 25 -98% hatching  
Refrigerator (10 days storage) 0-20% hatching  
15 and 20 days storage in refrigerator and BOD resulted in 0 % hatching
- Storage in Refrigerator (9°C)  
1 week storage 14 to 24% hatching  
2 to 5 weeks no hatching
- Laing and Eden (1990) reported viability of *Sitotroga cerealella* eggs for one month at 9°C



Means with different alphabets are significantly different from each other at LSD = 0.05 SED = 5.90; LSD (0.05) = 13.61

Mean per cent parasitism of Eri silkworm eggs stored at 9°C for different periods

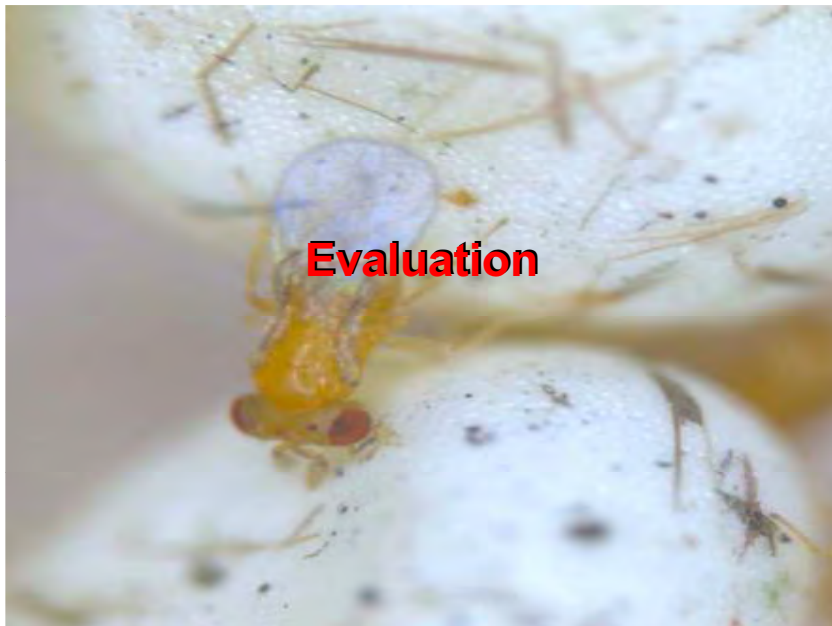
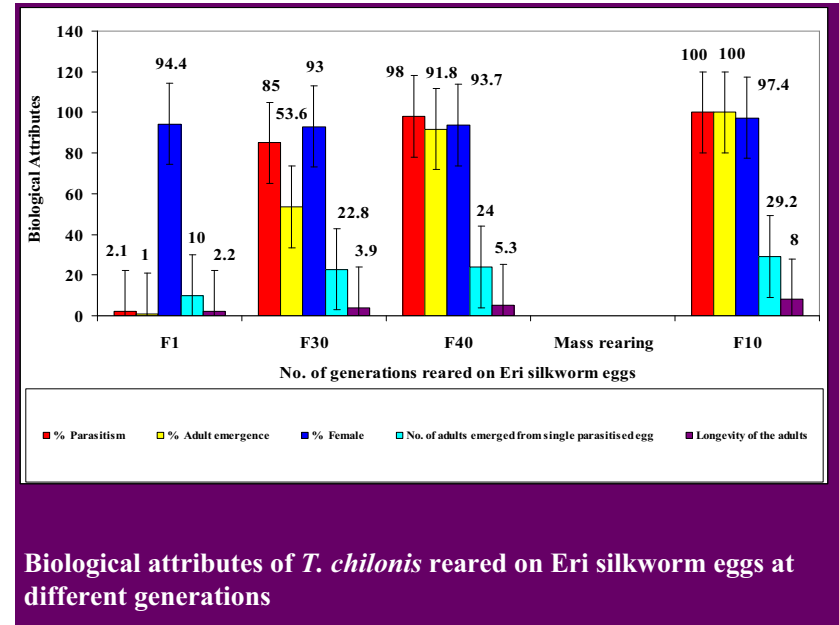




- ❑ Average developmental period: 9 -11 days
- ❑ Adult longevity: 1-8 days
- ❑ Per cent female progeny: 28.6 to 100
- ❑ Number of adults emerged from single parasitized egg: 7-34
- ❑ Per cent parasitism progressive: 2.1 to 98



- ❑ Nagarkatti *et al.* (1991) - frozen eggs of *Manduca sexta*, recorded 1- 20 eggs of *T. nubilale* from single parasitized egg
- ❑ Greenberg *et al.* (1996) - use of Eri silkworm eggs for rearing *T. chilonis*, *T. dendrolimi*, *T. cacoeciae*, *T. evanescens* and *T. pretiosum* & number of adults emerged from single parasitized egg ranged from 27- 60 with an average of 25



- Speed of travel (Distance 40cm)
  - Towards light (n = 17)
    - Time 2 to 4 min (Mean 3.2 min)
    - No. of adults 2 to 5 (Mean 3.4)
    - Per cent parasitism 41.0 to 79.7% (Mean 55.7%)
  - Opposite to light (n = 1)
    - Time 20 minutes
    - Parasitism 54.0%

❖ Host searching ability studied in 1 square foot cloth sleeved acrylic cage

❖ Influence of changing host (from Eri silkworm to *Corcyra*) on biological parameters of *T. chilonis*

**Host searching ability of *T. chilonis* reared on Eri silkworm eggs**

Treatments (No. of Parasitoids/cage)	Per cent parasitism of the single female on <i>Corcyra</i> eggs		Mean (B)
	<i>T. chilonis</i> reared on Eri silkworm eggs	<i>T. chilonis</i> reared on <i>Corcyra</i> eggs	
One female	59.8 <sup>a</sup>	29.6 <sup>b</sup>	44.7 <sup>a</sup>
Two females	36.4 <sup>b</sup>	25.4 <sup>b</sup>	30.9 <sup>a</sup>
Five adults	42.9 <sup>a</sup>	22.8 <sup>b</sup>	32.9 <sup>a</sup>
Ten adults	29.8 <sup>b*</sup>	34.3 <sup>b</sup>	32.1 <sup>a</sup>
Mean (A)	42.2 <sup>a</sup>	28.0 <sup>b</sup>	35.1

A Factor SED = 4.84 & LSD (0.05) = 9.86

B Factor SED = 6.84 & LSD (0.05) = 13.94

A x B Factor SED = 9.68 & LSD (0.05) = 19.71

Means with different alphabets are significantly different from each other at LSD = 0.05; \*- super parasitism was observed

**Influence of changing host on per cent parasitism of single female and per cent females emerged from parasitized eggs**

Generations	% parasitism of single female	% females emerged
Parent	37.9 <sup>b</sup>	49.4 <sup>b</sup>
F <sub>5</sub>	90.5 <sup>a</sup>	74.0 <sup>a</sup>
F <sub>10</sub>	83.5 <sup>a</sup>	74.6 <sup>a</sup>
SED	5.21	4.44
CD (0.05)	14.45	12.32

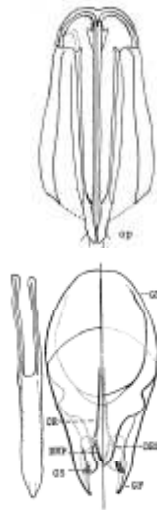
Means with different alphabets are significantly different from each other at LSD = 0.05

**Morphological parameters of *T. chilonis* reared on Eri silkworm and *Corcyra***

- ❖ Measured total body length, width, size of ovipositor / aedeagus at 50x magnification
- ❖ Data subjected to paired t - test & correlation between per cent parasitism and per cent females

**Morphological characters of *T. chilonis* reared on Eri silkworm and *Corcyra* eggs**

Character	Female reared on Eri	Female reared on <i>Corcyra</i>	Male reared on Eri	Male reared on <i>Corcyra</i>
Length mean	99.5	94.2	95.6	97.8
t- test	2.34*		-0.89	
Width mean	39.2	31.8	37.5	32.4
t-test	7.65***		3.86**	
Ovipositor /Aedeagus mean	44.3	38.9	43	39.3
t-test	5.24***		2.34*	



\* significant at 0.05; \*\* significant at 0.01 & \*\*\* significant at 0.001

**➤ Association of endo-symbiont in the adults of *T. chilonis* reared on Eri silkworm eggs**

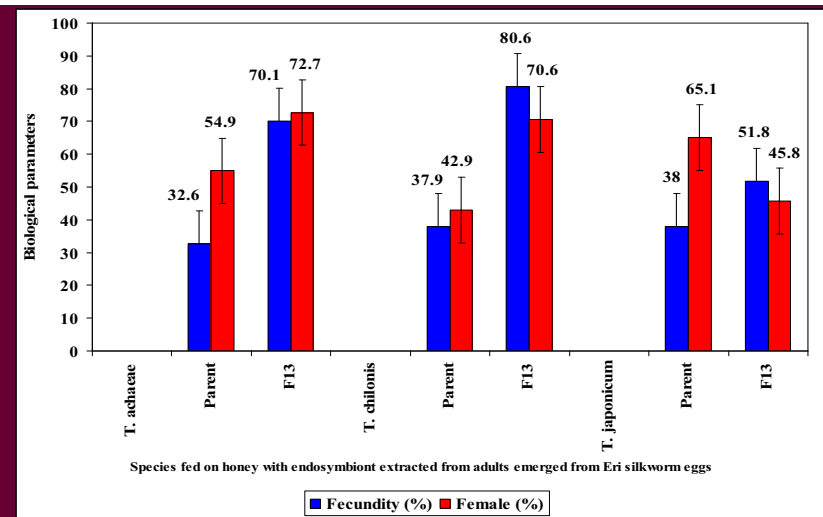
Adults killed, surface sterilized, processed, homogenized, spread on YEPD agar plates and incubated, Yeast colonies identified belonged to *Candida* group, single colony isolation

**➤ Influence of endo-symbiont on biological attributes of other species reared on *Corcyra* eggs**

Endo-symbiont yeast isolated grown on YEPD broth, pelleted colonies, mixed with honey and water at 1:1:2 (v/v/v) ratio, fed to laboratory reared *T. achaeae*, *T. chilonis* and *T. japonicum* for 13 generations, recorded fecundity and per cent females compared with parent



**Orange and white colonies of yeast isolated from *T. chilonis* reared on Eri silkworm eggs**



**Influence of endo-symbiont isolated from *T. chilonis* reared on Eri silkworm eggs on biological attributes on different species of *Trichogramma***

## Mass production & Cost Economics

### Mass production

After 10 generations of Mass rearing

Per cent parasitism: 100

Per cent females: 97.4

Number of adults from single parasitized egg: 29.2

Longevity of adults: 8 days



### Cost Economics

- Cost of production of Tricho cards required for release in one hectare @ 3 cards (ca. 18000 adults per card) US \$ 0.81

Cost of production reduced by 47.6% in comparison to the production using *Corcyra*



## Salient findings

- ❖ Storage of ES eggs at 9°C for 2 weeks completely prevented hatching, whereas UV sterilization required to prevent hatching in *Corcyra* eggs
- ❖ Initial per cent parasitism 2.1%, increased progressively to 100% after 10 generations of mass rearing
- ❖ 7-34 adults emerged from single parasitized ES egg, with 93 – 98% females, whereas on *Corcyra* generally only 1 adult emerges, with 40-50% females among progeny produced
- ❖ 83.5 to 90.5% parasitism and 74 to 74.6% females after shifting from rearing on ES to *Corcyra* eggs
- ❖ Endo-symbiont enhanced mean fecundity by 54%, 53% and 27% in *T. achaeae*, *T. chilonis* and *T. japonicum*, respectively, and mean female percentage by 25% and 39% in *T. achaeae* and *T. chilonis*, respectively
- ❖ Cost of production of Tricho card (utilizing ES eggs) required for release in one hectare @ 50,000/ha = US \$ 0.81; production cost reduced by 47.6% in comparison to that using *Corcyra* eggs

## Acknowledgements

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