

## Studies on Developing Races of *Trichogramma australicum* Girault Suitable for High Temperature - Low Humidity Conditions\*

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### ABSTRACT

Strains of *Trichogramma australicum* with tolerance to high temperatures (32-35 °C) could not be selected from a heterogeneous population of the parasite developed by interbreeding cultures obtained from Ambajipet, Cuddalore, Delhi, Lucknow, Ludhiana and Mandya. The Ludhiana, Delhi and Ambajipet cultures were relatively more tolerant to high temperature (35° C)-low humidity (10% RH). Significant improvements in adult emergence, fecundity and progeny production were attained after rearing the Ludhiana and Delhi cultures for 32-33 generations at progressively increasing temperatures from 30° to 33° C and decreasing humidities from 60 to 10%.

### INTRODUCTION

Successful utilization of *Trichogramma* spp. against different species of sugarcane borers would lead to the adoption of a suitable integrated control strategy against the pests. Extensive trials with indigenous *Trichogramma* parasites were conducted in India mainly for controlling the early shoot borer, *Chilo infuscatellus* Snellen. Periodic mass liberations of the parasite were either unsatisfactory or inconsistent in many cases (Gupta, 1951; Narayanan and Mookherji, 1953). The parasite was particularly found to be ineffective in northern sub-tropical belt during the hot months of April to June when extremes of high temperature and low humidity were experienced (Gupta,

1953, 1956). The present studies were undertaken to explore the feasibility of developing races of *Trichogramma australicum*, the indigenous species, that are tolerant to high temperature-low humidity conditions.

### MATERIALS AND METHODS

The parasites were collected from different regions in the country, viz. Ambajipet (A) in Andhra Pradesh, Cuddalore (C) in Tamil Nadu, Delhi (D), Lucknow (L1) Ludhiana (L2) and Mandya (M) in Karnataka. A part of the laboratory stock obtained from a particular source was mixed with field collected material from the same area and the cultures thus obtained were maintained individually. The original cultures were named after the places

\* Part of a thesis submitted by the senior author for the Ph.D. degree of the Post Graduate School, I.A.R.I., New Delhi-12, 1970.

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from where they were obtained. General morphological characters indicated that the material used in these studies belonged to the same species, *T. australicum*.

The cultures were maintained on *Corcyra cephalonica* eggs as the host. The bulk rearings were carried out at  $26 \pm 1.5^\circ\text{C}$  in desiccators maintaining 75% R. H. In order to reduce super parasitism to the minimum, care was taken to provide adequate number of host eggs for parasitisation and the egg cards were exposed to parasitisation only for a period of 12 hr. Inbred lines of different cultures were established by successive breeding from healthy mating pairs drawn out from the general collections. The physiological compatibility of the cultures was ascertained by a series of cross breeding studies. The populations belonging to the different cultures were pooled by putting together the *Corcyra* eggs parasitised by the E1 adults from the crosses A x C, A x L1, A x L2, A x M, C x D, C x L1, C x L2, C x M, D x L1, D x L2, D x M, L1 x L2, L1 x M and L2 x M and allowing the F2 adults to emerge and interbreed. These were then multiplied for another generation and the F3 adults were exposed to  $33^\circ\text{C}$  for a period of 4 generations and the resulting population was subsequently transferred to  $35^\circ\text{C}$  and reared for as many generations as possible. The performance of the resulting population at 33 and  $35^\circ\text{C}$  was compared to that of the Delhi culture which was kept as control.

The performance of the cultures under different temp.-humidity condi-

tions was studied from the duration of development, percentage adult emergence, adult longevity, sex-ratio, total reproductive period in females, fecundity and progeny production. To determine the longevity of adult parasites, the period elapsing between the time of their emergence and the time of death was considered separately for both sexes. This was accomplished by periodical observations of the pairs confined for oviposition. Fecundity was studied by taking counts of *Corcyra* eggs turning black as a result of parasite development. The total period during which the females laid eggs was recorded as the total reproductive period of the females. This was observed from egg cards removed at periodic intervals ranging from 5-10 hr. The total number of progeny emerging from eggs parasitised by a single female was recorded as progeny production from a single female. While taking counts of progeny produced from a single female, the individuals were sexed and the ratio of the total number of female to the total number of male estimated as sex-ratio. The duration of development was found out by calculating the weighted mean of the number of days taken by all the emerging adults belonging to both sexes. On the basis of parasites emerging out from them the percentage was calculated.

## RESULTS AND DISCUSSION

1. Screening for high temperature tolerance: The mean values of the characters studied at  $33^\circ$  and  $35^\circ\text{C}$  are given in Table I.

TABLE I. Performance of the pooled population obtained from the different cultures at 33° C and 35° C (at 75% RH) as shown by different characters.

Generation No.	Conditions of rearing		Adult emergence %	Adult longevity (hours)		Sex-ratio (Females/males)	Total reproductive period in females	Fecundity	Progeny production
	Temp.°C	RH%		Females	Males				
1	33	75	70.75	30.15	21.62	4.03	26.87	17.10	7.61
2	33	75	68.42	28.05	24.40	3.88	22.75	13.77	9.07
3	33	75	70.22	32.45	21.87	3.81	23.10	15.67	8.20
4	33	75	73.72	31.50	20.45	3.98	25.92	15.20	7.20
5	35	75	22.77	21.82	13.85	3.25	14.02	4.32	2.12

The variability in performance of the resultant population and the Delhi culture which was directly exposed to the same conditions, was tested. There was no significant difference between the surviving pooled population and the Delhi culture at 33° C. But a significant reduction in adult emergence was detected when exposed to 35° C. The percentage adult emergence in the pooled stock was only 22.27 as compared to 40.12 in the control culture (Delhi). Further, the pooled population could not maintain itself at 35° C for more than a generation. The results of these experiments showed that hybridization and pooling of all the cultures was not effective in selecting out a population with increased temperature tolerance. It was also indicated that some of the six cultures with which the pool was formulated might be much inferior in performance under high temperatures to the Delhi culture.

II. Performance of the different cultures at 35°C and 10% R-H. The developmental period of the different

cultures ranged from 6.64 to 7.47, the variation being non-significant. Significant variability was detected among the different cultures with regard to the percentage adult emergence (table II). More adults emerged from the Ludhiana, Delhi and Ambajipet cultures (42.20, 40.67, 36.60% respectively). The adult emergence in the F2 Generation ranged from 11.70 to 48.80%. The females emerging out from the Delhi culture lived for 30.15 hours while those from the Mandya culture were very short lived (13.95 hours). The variability in adult male longevity was also found to be significant the range being from 18.35 hours in the Ludhiana culture to 11.70 hours in the Mandya culture. In the Ambajipet culture the sex-ratio was very high (9.44). In the Ludhiana, Delhi and Lucknow cultures the sex-ratios were 3.14, 2.76 and 2.70 respectively. The total reproductive period in females of the Delhi culture was 22.80 hours and in the rest of the cultures females showed significantly lower values ranging from 8.07 to 14.42 hours only. The mean fecundity was observed to be 6.80 in the

TABLE II: Mean values of the different characters of the different cultures when reared at 35°C and 10% R.H.

Characters studied	Mean values for different cultures						Significance	C. D. (P=0.01)
	A	C	D	L1	L2	M		
Developmental period (days)	7.47	6.92	7.12	7.38	6.63	7.34	N.S.	—
Percentage adult emergence	36.60	18.25	30.67	14.72	42.20	20.12	**	9.144
Adult longevity (Female) (hours)	22.87	20.07	30.15	14.05	25.35	13.95	**	8.201
Adult longevity (male) (hours)	14.82	14.30	16.35	12.10	18.35	11.70	**	6.205
Sex-ratio (Female/male)	9.44	7.20	2.76	2.70	3.14	5.48	**	1.146
Total reproductive period in females (hours)	11.27	10.35	22.80	8.07	14.42	8.62	**	6.539
Fecundity (No. of eggs/female)	4.22	0.50	5.42	0.30	6.80	2.30	**	3.618
Progeny production per female	0.77	0.00	2.65	0.00	2.02	0.27	**	1.685
Percentage F <sub>2</sub> emergence	18.84	—	48.80	—	29.68	11.07	—	—
Percentage adults with malformed wings (Females)	74.37	74.25	69.22	75.47	69.85	68.25	N.S.	—
—Do— (Males)	36.77	70.12	58.10	66.15	47.75	74.75	**	11.800

\* Estimated from the F1 population. \*\* Significant at 1% level. N.S. Non-significant.

Ludhiana culture while the lowest value was recorded in the Lucknow culture (0.30). The progeny production was suppressed in the Lucknow and Cuddalore cultures. The F1 progeny produced by the Delhi and Ludhiana cultures were 2.65 and 2.02 per female and these were distinctly higher than in the other cultures.

Maximum number of malformed adult males emerged from the Mandya culture (74.75%) and in the Ludhiana and Ambajipet cultures, these were significantly lower being 47.75 and 36.77 per cent respectively. Sharma (1968) reported that the percentage of malformed adults ('runts') was more in females under conditions promoting superparasitism. In these experiments since all precautions were taken to avoid superparasitism, the appearance

of 'runts' seems to be due to adverse temperature condition. Goldschmidt (1938) reported that in *Drosophila melanogaster* Heign, changes in shape of wings occurred as a result of exposure to heat shock. It will be seen that Ludhiana, Delhi and Ambajipet cultures were found to be relatively tolerant to high temperature and low humidity conditions as evidenced by the percentage adult emergence and overall better performance. Schepetnikova (1939) observed that oviposition of *T. evanescens* was suppressed at 35°C. This may perhaps be explained on the basis of the fact that the area in which these experiments were conducted was of a cooler climate and a temperature of 35°C was therefore extremely adverse to the indigenous population. The increased tolerance of the Delhi, Ludhiana and Ambajipet

cultures to 35°C and 10 per cent R. H. can be normally expected since these areas, particularly Delhi and Ludhiana are characterised by extremes of high temperatures and low humidity during summer months of May-July. It is likely that under these conditions natural selection operative on the population resulted in increased tolerance to these adverse conditions.

The Lucknow and Cuddalore cultures showed high susceptibility to the temperature-humidity stress conditions. The absence of a high temperature tolerance in the Lucknow culture obtained from a region with similar climatic conditions as in Delhi and Ludhiana cultures appeared to be somewhat unexpected. It is understood that the Indian Institute of Sugarcane Research Lucknow has been obtaining cultures from elsewhere and releasing them in their environments. Obviously the population has not undergone any appreciable natural selection for high temperature tolerance.

**III. Selection of the cultures for high temperature - low humidity Conditions :** In view of the better performance of the Ludhiana, Delhi and Ambajipet cultures at 35°C and 10 per cent R.H. efforts were made to improve their performance under temperature-humidity stress conditions by selective breeding. This was done by rearing these cultures at increasing temperatures from 30° to 33° C and at decreasing humidity levels (60 to 10 per cent R.H.).

A part of the culture reared at 10 per cent R.H. and 33° C was main-

tained for four subsequent generations at the same conditions to test for consistency of performance. The rest of the culture was exposed to 35° C and 75 per cent R. H. The relative humidity at this temperature was kept at a near optimum level in order to prevent loss or extinction of the culture due to high temperature-low humidity stresses given simultaneously. The Ludhiana, Delhi and Ambajipet cultures reared at progressively adverse conditions would henceforth be referred to as the 'P' cultures and the corresponding control cultures as 'Q' series, i. e., directly exposed to the particular conditions from stocks held at  $26^{\circ} \pm 1.5^{\circ}$  C.

The performance of the 'P' cultures was found to be more or less similar at progressively adverse temperature-humidity conditions. When the temperature was increased from 30° to 33° C and humidity decreased from 60 to 10%, all the 'P' stocks revealed decline in adult emergence, fecundity and progeny production, but the extent of decline of these characters in the 'P' cultures was not as large as that observed for the corresponding 'Q' cultures. At all stages of rearing under progressively adverse conditions the 'P' cultures showed higher mean values for the characters tested.

The differences in the 'P' and 'Q' Ludhiana cultures were significant at 33° C and 10 per cent R.H. at 34th generation, in respect of fecundity and progeny production. These improvements were maintained even in the 38th generation at the same set of conditions. This revealed consistency

in the improvements attained as a result of selective breeding. The improvement in adult emergence at 33°C and 10 per cent R.H. attained by selective breeding of the Delhi 'P' culture was significant and consistent in respect of adult emergence. Significant differences in the P and Q cultures of Ambajipet were not evident at 33°C in respect of fecundity, progeny production and adult emergence. All the 'P' cultures from Ludhiana, Delhi and Ambajipet stock when brought to 35°C and 75 per cent R.H. registered improvements in adult emergence, fecundity or progeny production. The 'P' lines in the Delhi and Ludhiana stocks showed improvements in fecundity, progeny production and adult emergence. The Ambajipet 'P' culture registered improvement in adult emergence and progeny production. But the 'P' cultures in the Ambajipet and Delhi stocks could not be maintained beyond two generations at 35°C and 75 per cent R.H. The Ludhiana culture could however be maintained for three generations. Obviously, the extinction of the selected cultures at 35°C is due to the progressively deleterious effect of the very same adverse temperature condition. The increase in tolerance to temperature-humidity stress conditions is explicable on the basis of elimination of susceptible genotypes and the selection of those which are more tolerant to these conditions.

Wilkes (1942) developed cold tolerant strains of *Dahlbominus fuliginosus* by releasing the adults in a temperature gradient and by selection of those individuals which congregated

in the 6° to 10° C zone. When selection was thus continued for four generations a strain could be developed in which over 50 per cent of the adults preferred temperatures below 12.5° C. In these studies, the selection of individuals with the required preferendum could be directly made and this explains the rapidity with which cold tolerant strains could be developed. In the present experiments since the selection pressure viz., high temperature-low humidity conditions was purposely kept low so as to avoid extinction of the cultures, the relatively slow process of selection could be expected. Sharma (1968) reported that low humidity tolerance in *T. evanescens minutum* could be improved by rearing the parasite at 30° C for 10 generations each at 75, 60, 45, 30 and 15 per cent R. H. In these experiments also selection pressure was not rigorously applied and it is seen that the process of selection was relatively slow. However, a comparison of these results with those reported in the present paper is not possible in view of the variation of the nature and extent of the selection pressures applied. The results indicate that in Ludhiana, Delhi and Ambajipet cultures which appeared to be relatively tolerant to high temperature - low humidity conditions, the biotic potential could be increased by rearing in progressively adverse temperature - humidity combinations.

#### ACKNOWLEDGEMENTS

The authors are grateful to the Director, Indian Institute of Sugarcane Research, Lucknow, the Head of

the Division of Zoology - Entomology, Punjab Agricultural University, Ludhiana, the Assistant Entomologists, Parasite Breeding Stations at Ambajipet and Mandya, and the Sugarcane Specialist, Sugarcane Research Station, Cuddalore for supplying parasite cultures and for providing necessary facilities for making field collections.

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