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Evaluation of Interspecific Interaction Between *Encarsia lutea* and *Eretomocerus near mundus*, the Parasitoids of Cotton Whitefly (*Bemisia tabaci*) with Host Plants Density Levels and Preference

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Abstract: Studies were carried out at CABI Biosciences Pakistan Centre, Rawalpindi to evaluate the effects of host plants and insects with host density levels and preference by both these species, i.e., *Encarsia lutea* and *Eretomocerus* separately as well as together in competitive situation. Preference of density levels by both parasitoids species showed that density level 20 (cotton leaf disc with 20 *Bemisia tabaci* nymphs and pupae) was contributed more (42%) preference than other density levels (5, 10, 40, 60 and 80). Different density levels had highly significant effect on settlement time of both species separately while in competitive situation the settlement time of both species was not significantly different from each other on different treatments.

Key words: Parasitoids, density levels, *Bemisia tabaci*, settlement time and intergeneric

INTRODUCTION

When more than one parasitoid species seek to attack the same prey in a host population, the possibility of interspecific interaction between these species increases. Such interactions can lead to either displacement of one parasitoid by another or reduction in abundance of one species by the species other one. In some cases overall effectiveness of biological control of pests can remain unaffected, or even improved by the interaction while in others control is diminished (Price, 1975).

Interspecific competition is a very common phenomenon and can exert a major influence on the population size of different species in natural communities. Therefore, this study shows great importance. Food, mates and space are the possible source of competition for the parasitoids (Colinvaux, 1973).

Parasitoids are ecological homologous and cannot coexist in the same habitat unless they possess different alternative hosts or some other modifying factors. In the long run one species must loose out and disappear unless it involved some adoption to escape from competition. Competition for resources also restricts local distribution. Some species drive other out by aggressive interaction (DeBach, 1974).

Coexistence of parasitoid populations of different niches (Gause, 1934; Hardin, 1960) or that competing

species show an independent aggregated distribution (Atkinson and Shorrocks, 1984; Ives and May, 1985). Host specifying is one of the main factors resulting in niche segregation (Vet and Van Alphen, 1985).

According to McNaughton and Wolf (1973) there are four possible outcomes of interspecific competition in two species systems.

- One species is the better competitor and always wins.
- The other one is better competitor and always wins.
- The species coexist independently in a stable equilibrium.
- There is an unstable equilibrium although the eventual outcome is that one species or the other always wins.

Cotton is an important cash crop of Pakistan. Indiscriminate use of the pesticide on cotton has created a number of problems and has resulted in flare of *Helicoverpa* and *Bemisia tabaci*. Both of these have developed resistance against almost all the present day commonly used pesticides and have virtually become impossible to control with pesticides. Moreover, they do not give good control of *Bemisia tabaci* because of long incubation period and protective waxy covering on the nymph and pupae (Jhonson *et al.*, 1982). The intensive use of insecticides adversely affects natural enemies resulting in white fly higher infestation.

Pest complex attacking cotton can be divided into two groups, i.e., sucking and boring. Among sucking pest *Bemisia tabaci* is the most injurious pest damaging cotton crop by sucking cell sap and transmitting cotton Leaf curl virus (CLCV). Whitefly (*Bemisia tabaci*) is wide spread in Asia, Africa, America, Russia, Australia and the Pacific Islands (Mohyuddin *et al.*, 1985). The use of pesticides can greatly be reduced by developing Integrated Pest Management program (IPM). In the IPM, biological control is the main component of the pest control and other methods fit into this because biological control of a particular pest species already exists in the agro-ecosystem and this should not be disturbed (Falcon and Smith, 1973). Therefore this study shows great importance.

Eleven species of parasitoids belonging to aphelinid have been recorded from the nymph/pupae of whitefly by the Pakistan Agricultural Research Council (PARC)/ International Integrated Biological Control (IIBC) station and Centre for Agriculture and Biosciences (CAB international institute of biological control). Of these *Encarsia lutea* and *Eretomocerus near mundus* are of significant importance. Studies were carried out to evaluate the interspecific interaction between these two parasitoid species to determine the efficacy of parasitoids as effective biocontrol agent. Keeping in view of the above stated factors the studies of intergeneric interaction between *Encarsia lutea* and *Eretomocerus near mundus*, the parasitoids of cotton whitefly (*B. tabaci*) were initiated.

MATERIALS AND METHODS

To evaluate intergeneric interaction between *Encarsia lutea* and *Eretomocerus near mundus*, three studies were carried out at CABI Biosciences Pakistan center, Rawalpindi. Methods for each study are described as: Host density preference by *Encarsia lutea* and *Eretomocerus near mundus* when released alone and together on excise leaf discs of cotton, brinjal and okra containing *Bemisia tabaci* nymphs and pupae:

To determine the preference of different density levels of *B. tabaci* by two parasitoid species, landing time, number of parasitoids landing and settlement time were taken into consideration. Free choice test was conducted in a glass cage having 1×1.5 ft dimension. The following 8 treatments were used in completely randomized design as (i) cotton leaf disc with five *B. tabaci*, nymphs and pupae, (ii) Cotton leaf disc with ten *B. tabaci*, nymphs and pupae, (iii) Cotton leaf disc with twenty *B. tabaci*, nymphs and pupae, (iv) Cotton leaf disc with forty *B. tabaci*, nymphs and pupae, (v) Cotton leaf disc with sixty

B. tabaci, nymphs and pupae, (vi) Cotton leaf disc with seventy *B. tabaci*, nymphs and pupae, (vii) Cotton leaf disc with eighty *B. tabaci*, nymphs and pupae, (viii) Cotton leaf disc without any host i.e., whitefly and (ix) Empty petridish as control. One mated female of each parasitoid species were released in cages separately and two mated females simultaneously. There were thirty replications for this study.

Statistical analysis: The data were subjected to ANOVA and treatment means were compared by the LSD test at alpha 0.05, free test method and chi square analysis (Steel *et al.*, 1997).

RESULTS AND DISCUSSION

Host density finding by *Encarsia lutea* and *Eretomocerus near mundus* when released alone and together on excise leaf discs of cotton, brinjal and okra.

In order to assess the effect of host density on host finding behavior by the two parasitoid species data on 3 variables were collected.

The number of parasitoids landing on different host density levels: Assuming significant effect of treatments, the variance of observed frequencies from the expected frequencies provided analysis for preference (Table 1). Number of *E. lutea* females landing was significantly different ($\chi^2 = 17.4$) at $\alpha = 0.05$ on different density levels. Host density levels seemed to effect host finding by *E. lutea* as significantly higher number of female adult (9 observed in comparison with 3.75 expected) landed on density level 20 (T_3) as compared with other density levels. Of the total ($\chi^2 = 17.4$) more than 42% was contributed by density level 20 alone indicating more preference (Table 1). Similarly, the number of mated *E. mundus* landing on different host plants densities was significantly ($\chi^2 = 16.1$, $\alpha = 0.05$.) different when exposed alone. Density level 20 contributed maximum (45%) from the total ($\chi^2 = 16.1$) indicating maximum preference than other density levels (Table 2).

Highest number (i.e., 14) of parasitoids landed on density level 20 while control (empty petri dish) was least attracted (Table 1 and 3). Selection of hosts by two species seems to be positively correlated on different density levels. There was a significant trend for both species to coexist on the same treatment ($\chi^2 = 67.8$ for T_1 , $\chi^2 = 29.1$ for T_2 , $\chi^2 = 13.4$ for T_3 , $\chi^2 = 23.8$ for T_4 , $\chi^2 = 67.8$ for T_5 , $\chi^2 = 67.8$ for T_6 , $\chi^2 = 82.2$ for T_7 , $\chi^2 = 54.4$ for T_8), respectively (Table 3).

The correlation of both species on different treatments was positive as measured by Cole's index.

Table 1: Host density level finding by *Encarsia lutea* for parasitism of *Bemisia tabaci* (nymphs and pupae) in glass cage in free choice test for 20 min observation time when released alone

Treatments	Different density level	Observed frequency	Expected frequency	χ^2	Contribution to χ^2 (%)
T ₁	5	3	3.75	0.15	00.86
T ₂	10	3	3.75	0.15	00.86
T ₃	20	9	3.75	7.35	42.20
T ₄	40	8	3.75	4.81	27.60
T ₅	60	1	3.75	2.01	11.55
T ₆	80	3	3.75	0.15	00.86
T ₇	Leaf without host	1	3.75	2.01	11.55
T ₈	Empty petridish	2	3.75	0.81	04.60
	Total: 8	30	30.00	17.40*	99.90

*Significant at p = 0.05, df = 7

Table 2: Host density level finding by *E. near mundus* for parasitism of *Bemisia tabaci* (nymphs and pupae) in glass cage in free choice test for 20 min observation time when released alone

Treatments	Different density level	Observed frequency	Expected frequency	χ^2	Contribution to χ^2 (%)
T ₁	5	1	3.75	0.81	05.03
T ₂	10	8	3.75	4.81	29.80
T ₃	20	9	3.75	7.35	45.60
T ₄	40	4	3.75	0.01	00.06
T ₅	60	2	3.75	0.81	05.03
T ₆	80	3	3.75	0.15	00.93
T ₇	Leaf without host	3	3.75	0.15	00.93
T ₈	Empty petridish	0	3.75	2.01	12.40
	Total: 8	30	30.00	16.10*	99.70

*Significant at p = 0.05, df = 7

Table 3: No. of *Encarsia lutea* and *Eretomocerus near mundus*, adult females landing on *Bemisia tabaci* colonies on three host plants and empty petridish in glass cage 20 min observations time

Parasitoid exposed	5			10			20			40			60			80			LWH			EPD		
	OF	EF	χ^2	OF	EF	χ^2	OF	EF	χ^2	OF	EF	χ^2	OF	EF	χ^2	OF	EF	χ^2	OF	EF	χ^2	OF	EF	χ^2
A	0	7.5	7.5	2	7.5	4.7	3	7.5	2.70	3	7.5	2.7	0	7.5	7.5	0	7.5	7.5	0	7.5	7.5	2	7.5	4.00
B	2	7.5	4.0	2	7.5	4.7	6	7.5	0.30	5	7.5	0.8	2	7.5	4.0	2	7.5	4.0	0	7.5	7.5	1	7.5	5.60
AB	1	7.5	5.6	6	7.5	0.3	5	7.5	0.80	3	7.5	2.7	1	7.5	5.6	1	7.5	5.6	1	7.5	5.6	2	7.5	4.00
C	27	7.5	50.7	20	7.5	20.8	16	7.5	9.60	19	7.5	17.6	27	7.5	50.7	27	7.5	50.7	29	7.5	61.6	25	7.5	40.8
Total	30	30.0	67.8*	30	30.0	29.1*	30	30.0	13.43*	30	30.0	23.8*	30	30.0	67.8*	30	30.0	67.8*	30	30.0	82.2*	30	30.0	54.4*

*Significant at p = 0.05, df = 3, OF = Observed Frequency, EF = Expected Frequency, A = No. of *E. lutea* present alone, B = No. of *E. near mundus* present alone, AB = No. of both species present, C = No. of both species present, LWH = Leaf discs without hosts, EPD = Empty petridish

The co-relations between two species as measured by co-relation co-efficient seems to be associated with each other slightly positively on T₂ (V = 0.206 Var (V) = 0.039), T₃ (V = 0.209 Var (V) = 0.145), T₄ (V = 0.263, Var (V) = 0.159), as presented in Table 4.

Time spent by parasitoid female before landing on different density levels after release (landing time):

Analysis of variance for landing time *E. lutea* and *E. mundus* when released alone on different density levels was not significant (F = 1.26, Pr>F = 0.315, F = 1.04, Pr>F = 0.435) as given in appendices 1 and 2. The mean landing time on different density level was not significantly different from each other and the mean landing time in minutes of *E. lutea* on host density level was (\bar{x} = 7.5) host density level 10 (\bar{x} = 6.3) host density level 20 (\bar{x} = 6.3) host density level 40 (\bar{x} = 7.6), host density level 60 (\bar{x} = 9.0), host density level 80 (\bar{x} = 2.0), leaf without host density (\bar{x} = 2.0), empty Petri dish (\bar{x} = 6.0), respectively (Table 5). Mean landing time

of *E. mundus* on different treatments was on destiny level 5 (\bar{x} = 5.0) host density level 10 (\bar{x} = 3.9) host level 20 (\bar{x} = 5.7) host density level 40 (\bar{x} = 3.7) host density level 60 (\bar{x} = 6.0) density level 80 (\bar{x} = 9.0) leaf without host density (\bar{x} = 5.3), empty Petri dish (\bar{x} = 0.6), respectively as shown in Table 5.

The preference two different levels of *B. tabaci* by both the species in competitive situation was not significant (F = 1.26 Pr>F = 0.3 for *E. lutea* and F = 1.25 Pr>F = 0.318 for *E. mundus*) as presented in appendices 4 and 5. The mean landing time on density level 5 was (\bar{x} = 1.4, \bar{x} = 14), density level 10 (\bar{x} = 7.0, \bar{x} = 6.7), density level 20 (\bar{x} = 5.2, \bar{x} = 5.7), density level 40 (\bar{x} = 5.2, \bar{x} = 5.5), density level 60 (\bar{x} = 5.0, \bar{x} = 6.0), density level 80 (\bar{x} = 4.5, \bar{x} = 9.0), leaf without host density level (\bar{x} = 0, \bar{x} = 4.0) empty Petri dish (\bar{x} = 0.9, \bar{x} = 5.3), for *E. lutea* and *E. mundus*, respectively indicating that one species influence the landing time of others (Table 6).

Time spent by *Encarsia lutea* and *Eretomocerus near mundus* on different density levels of *Bermisia tabaci* (settlement time): The analysis of variance for time spent by *E. lutea* on different density levels when released in cage alone is given in Appendix 1. The time spent by parasitoid female was not significantly different on different density levels ($F = 1.48$ $Pr > F = 0.225$, Appendix 2). The adult female of *E. lutea* spend maximum time in minutes (2.08) on density level 20 (Table 5). Analysis of variance for time spends by *E. mundus* is presented in Appendix 3 Different density levels had highly significant effect on settlement time of *E. mundus* ($F = 5.25$, $Pr > F = 0.001$). The mean settlement time in minutes of *E. mundus* was highest on density level 80 ($\bar{x} = 3.8$) and lowest on empty Petri dish ($\bar{x} = 0.1$) as shown in Table 5 (Appendix 4 and 5).

The analysis of variance for settlement time of both species, on different density levels in competitive situation is presented in Appendix 6 and 7. Time spent by *E. lutea* on different treatment was not significantly different ($F = 1.06$, $Pr > F = 0.621$) while the time spent by *E. mundus* was highly significant ($F = 3.91$) $pr > F = 0.066$) in competitive situations. The mean settlement time in minutes by *E. lutea* and *E. mundus* was on density level 5 ($\bar{x} = 1.2$, $\bar{x} = 1.4$) density level 10 ($\bar{x} = 1.2$, $\bar{x} = 1.6$) density level 20 ($\bar{x} = 1.6$, $\bar{x} = 0.9$), density level 40 ($\bar{x} = 1.4$, $\bar{x} = 1.0$) density level 60 ($\bar{x} = 2.0$, $\bar{x} = 0.9$) density level 80 ($\bar{x} = 0.7$, $\bar{x} = 2.0$), leaf without host density level ($\bar{x} = 0$, $\bar{x} = 2.5$) and empty peridish ($\bar{x} = 1.0$, $\bar{x} = 0.75$), respectively indicating that settlement time of both species was not significantly different from each other on different treatments in competitive situation (Table 6).

Table 4: Association of *E. lutea* and *E. nr. mundus* on different host density levels in a free choice test in glass cage for 20 min observation time at $p = 0.05$, $df = 7$

Treatments	Different level of host density	χ^2 Observed	χ^2 Tabulated	Cole's index		Point correlation coefficient	
				C1	C2	V	Var (V)
T ₁	5	0.07	14.02	-	-0.001	-0.049	0.0009
T ₂	10	1.28	14.02	0.46	-	0.206	0.0396
T ₃	20	0.29	14.02	0.25	-	0.209	0.0145
T ₄	40	2.08	14.02	0.87	-	0.263	0.1590
T ₅	60	0.07	14.02	-	-0.002	-0.049	-0.0149
T ₆	80	0.07	14.02	-	-0.002	-0.049	0.0149
T ₇	(LWH) Leaf with out host	0.00	14.02	0.00	0.000	0.000	0.0000
T ₈	Empty petridish (EPD)	0.27	14.02	1.00	-	0.522	-0.0402

Table 5: Preference of *Encarsia lutea* and *Eretomocerus nr. mundus* as indicated in landing and settlement times (min) of females released in a glass cage on different *Bemisia tabaci* density levels on mean basis

Treatments	Different level of host density	<i>Encarsia lutea</i>		<i>Eretomocerus nr. mundus</i>	
		Landing time	Settlement time	Landing time	Settlement time
T ₁	5	7.50a	1.70a	5.00ab	0.50c
T ₂	10	6.30a	1.50a	3.90ab	0.90c
T ₃	20	6.30a	2.08a	5.70ab	1.90b
T ₄	40	7.60a	1.80a	3.70ab	0.80c
T ₅	60	9.00a	1.60a	6.00ab	2.20ab
T ₆	80	2.00a	1.20a	9.00a	3.80a
T ₇	(LWH) Leaf without host	2.00a	0.90a	5.30ab	0.40bc
T ₈	(EPD) Empty petridish	6.00a	0.90a	0.60c	0.10c

Means of same letter(s) within a group are not significantly ($p > 0.05$) different by DMR test

Table 6: Preference of *Encarsia lutea* and *Eretomocerus nr. mundus* as indicated in landing and time spent (min) on treatments by females computing for host in a glass cage on different *Bemisia tabaci* density levels (mean value basis)

Treatments	Different level of host density	<i>Encarsia lutea</i>		<i>Eretomocerus nr. mundus</i>	
		Landing time	Settlement time	Landing time	Settlement time
T ₁	5	1.40a	1.20ab	14.00a	1.40ab
T ₂	10	7.00a	1.20ab	6.70bc	1.00ab
T ₃	20	5.20ab	1.60ab	5.70c	0.90c
T ₄	40	5.20ab	1.40ab	5.50c	1.00ab
T ₅	60	5.00ab	2.00a	6.00bc	0.90c
T ₆	80	4.50ab	0.70c	9.00b	2.00a
T ₇	(LWH) Leaf without host	0.00	0.00	4.00c	2.50a
T ₈	(EPD) Empty Petridish	0.90c	1.00ab	5.30c	0.75c

Means of same letter(s) within a group are not significantly ($p > 0.05$) different by DMR test

Appendix 1: Analysis of variance for settlement time of *E. lutea* on different density level conditions (5, 10, 20, 40, 60, 80, leaf without host, empty petridish)

Source	df	SS	MS	F-value	Pr>F
Type 1 SS					
Treatments	7	125.2060	17.6688	1.26	0.3153
Error	22	312.7600	14.2163		
Corrected total	29	437.9666			

Appendix 2: Analysis of variance for landing time of *E. nr. mundus* on different density levels

Source	df	SS	MS	F-value	Pr>F
Type 1 SS					
Treatments	7	88.720	12.675	1.04	0.435
Error	22	269.402	12.245		
Corrected total	29	358.130			

Appendix 3: Analysis of variance for settlement time of *E. nr. mundus* on different density levels during the exposure of both species

Source	df	SS	MS	F-value	Pr>F
Type 1 SS					
Treatments	7	30.041	4.290	5.25	0.0012
Error	22	17.968	0.816		
Corrected total	29	48.009			

Appendix 4: Analysis of variance for landing time of *E. lutea* on different density levels when exposed together

Source	df	SS	MS	F-value	Pr>F
Treatments	6	51.7080	8.618	1.26	0.314
Error	23	157.5000	6.847		
Corrected total	29	209.2096			

Appendix 5: Analysis of variance for landing time of *E. lutea* on different density levels when exposed both species exposed together

Source	df	SS	MS	F-value	Pr>F
Treatments	6	3.039	0.506	1.25	0.318
Error	23	0.328	0.405		
Corrected total	29	12.368			

Appendix 6: Analysis of variance for settlement time of *E. nr. mundus* on different density levels when both species exposed together

Source	df	SS	MS	F-value	Pr>F
Treatments	7	83.1950	11.880	1.06	0.421
Error	22	247.1700	11.235		
Corrected total	29	330.3660			

Appendix 7: Analysis of variance for settlement time of *E. nr. mundus* on different density levels when both species exposed together

Source	df	SS	MS	F-value	Pr>F
Treatments	7	3.769	0.538	3.91	0.0065
Error	22	3.028	0.137		
Corrected total	29	6.798			

df : Degree of freedom; SS: Sum of Squares; MS: Mean Squares

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