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# Effects of female mating status and age on fecundity, longevity and sex ratio in *Trichogramma minutum* (Hymenoptera: Trichogrammatidae)

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#### **ABSTRACT**

Effects of female mating status and age of *Trichogramma minutum* Riley on its fecundity, longevity and offspring sex ratio were determined in the laboratory, using eggs of the variegated cutworm as hosts. Although the mating status of female T, minutum did not affect their total fecundity significantly (P > 0.05), mated and unmated females showed different allocations of progeny. Mated females deposited significantly more eggs (P < 0.05) than those unmated on the first day of exposure to hosts. On subsequent days, however, unmated females parasitized significantly more hosts (P < 0.05) than those mated. Mated females laid 82.4% of their total fecundity on the first day of oviposition, whereas unmated females laid 58.3%. The number of eggs parasitized by both groups of females decreased significantly (P < 0.05) with parasitoid age. Unmated females lived longer (P < 0.05) than their mated counterparts. No significant differences (P > 0.05) in clutch size (the number of parasitoid offspring produced per parasitized host) and emergence rate were found between the offspring of mated and unmated female parasitoids. The sex ratio of the offspring of mated females changed significantly (P < 0.05) with maternal age: younger females produced a higher proportion of daughters than did older parasitoids. Unmated females produced male offspring only.

#### INTRODUCTION

The effects of host size (Stinner et al. 1974; Southard et al. 1982; Bai et al. 1992), rearing temperatures (Smith and Hubbes 1986; Jalali and Singh 1992), and food availability to females (Bai et al. 1992) upon reproductive potential of *Trichogramma* have been well documented. However, maternal mating status and age may influence longevity, fecundity and sex allocation in *Trichogramma*. Although Partridge (1986) reported that mated females of insects are generally shorter lived than virgins, the effect of maternal mating on reproductive potential of *Trichogramma* remains uncertain. Lund (1938) observed that there were no differences in longevity between mated and unmated females of *Trichogramma evanescens* Westwood, but there was a significant difference in fecundity. In contrast, Yu et al. (1984) found that longevity in mated and unmated *Trichogramma minutum* Riley differed significantly, but there was no significant difference in fecundity.

Reduced fecundity with maternal age has been documented in various insect species. However, a recent study by Navasero and Elzen (1992) demonstrated that the clutch size in mated *Microplitis croceipes* (Cresson) (Hymenoptera: Braconidae) varied cyclically over their life spans with egg production peaking in intermediately-aged parasitoids. Previous studies have shown that offspring sex ratio in *Trichogramma* is affected by maternal age. In studies on *T. minutum*, Houseweart *et al.* (1983) and Smith and Hubbes (1986) both reported that young females produced a higher proportion of female offspring than old females.

Although reproductive biology of *T. minutum* associated with other hosts has been studied (Yu et al. 1984; Smith and Hubbes 1986), no reports have been documented on this species with the variegated cutworm, *Peridroma saucia* (Hübner), a minor pest on field crops in British Columbia. In recent years, however, *P. saucia* becomes more and more abundant on small fruit crops. An ongoing field release of *Trichogramma* to control *P. saucia* on raspberry is currently conducting in the Fraser Valley (Henderson *et al.* unpub. data). The reproductive characteristics of commercial *T. minutum* with *P. saucia* may be different from that of field collected parasitoids with other hosts studied by others (Yu *et al.* 1984). The objectives of this study were to investigate the effects of female mating status on progeny allocation, longevity and fecundity in a commercial strain of *T. minutum*; and to determine sex allocation over the lifespan of mated female *T. minutum*, using eggs of *P. saucia* as hosts.

# **MATERIALS AND METHODS**

#### Host eggs.

The host eggs used in this study were obtained from a laboratory culture of *P. saucia* and were less than 24 h old.

#### Parasitoids.

Trichogramma minutum was obtained from a commercial source, reared on eggs of the Mediterranean flour moth, Ephestia (= Anagasta) kuehniella Zeller, and then reared in the laboratory on eggs of P. saucia for four generations prior to this study at  $21 \pm 2^{\circ}$ C,  $50 \pm 10\%$  RH, and under 14L:10D photoperiod.

Parasitized *P. saucia* eggs were isolated individually in gelatin capsules (20mm  $\times$  5 mm) to obtain individual parasitoids for the following experiments. Only newly ( $\leq$  3 h old) and singly eclosed individuals (one parasitoid developed from each parasitized host) were used to prevent any age and size factors of the parasitoids from influencing the results. Mated females were obtained by placing two pairs of virgin females and males in a gelatin capsule (20 mm  $\times$  5 mm) for 3-4 h. Because mating among virgin *Trichogramma* adults occurs readily (Nagarkatti and Nagaraja 1978), it was expected that all females would be inseminated in such a situation. Both mated and unmated females were unfed.

#### Fecundity and longevity.

Each of the mated and unmated females was transferred individually into a clear plastic Petri dish (50 mm X 9 mm) containing 75 host eggs. Twenty-four hours following introduction, each parasitoid was transferred into a second Petri dish containing 60 host eggs. The host eggs (=60)

were then changed every second day until the T. minutum died. Each female was a replicate and each of two treatment groups (mated and unmated) contained initially 59 females (= replicates). The experiments were conducted at  $25 \pm 1^{\circ}$ C and  $60 \pm 10\%$  RH with a 16L:8D photoperiod. Following incubation for 7 days under the above conditions, the parasitized eggs were counted using a dissecting microscope at 15X. To determine longevity, the parasitoids from the above experiment were observed at 8 h intervals until they died. Fecundity of T. minutum was evaluated as the average number of parasitized host eggs per female, and longevity as the average lifespan in hours.

# Clutch size, emergence rate and sex ratio of F1 progeny.

A maximum of 20 parasitized host eggs per Petri dish were selected randomly and incubated until adult parasitoids emerged. The numbers of male and female progeny and the uneclosed parasitized eggs were counted. Uneclosed parasitized eggs were individually dissected to determine the number of developing parasitoid offspring. Clutch size (the number of parasitoid offspring produced per parasitized host) and emergence rate were calculated as:

$$Clutch size = \frac{total\ eclosed\ progeny + total\ uneclosed\ progeny}{total\ parasitized\ host\ eggs\ incubated}$$

Emergence rate = 
$$\frac{\text{total eclosed parasitized eggs}}{\text{total parasitized eggs incubated}} \times 100\%$$

*Trichogramma minutum* is an arrhenotokous species in which virgin females produce only male offspring, whereas mated females produce both male and female progeny. Therefore, the sex ratio of F1 progeny was only comparable among age groups of the mated females.

# Data analyses.

The data were transformed as either  $\arcsin \sqrt{P}$  or  $\sqrt{x+0.5}$  before ANOVA (Zar 1984), where P represents the percentage of emergence rate or sex ratio, and x is mean number of the parasitized eggs, clutch size, or longevity. One-way ANOVA was used to estimate significances. Significant differences were separated by Duncan's multiple range test at P = 0.05 level.

#### RESULTS AND DISCUSSION

### Fecundity and longevity.

Mating status of female parasitoids significantly affected their daily fecundity (F = 15.34; df = 1, 116; P = 0.0002) (Table 1). Mated females parasitized more host eggs than unmated counterparts on the first day of oviposition, but fewer on subsequent days. The results suggest that mating stimulates female T. minutum to deposit eggs quickly. The fecundity of T. minutum on the first day of emergence in this study was greater than that reported by Yu et al. (1984) and Smith and Hubbes (1986). Although mated and unmated T. minutum followed different progeny allocation strategies in their lifetimes, their respective total fecundities per female were not significantly different (F = 0.23; df = 1, 116; P = 0.6355). Yu et al. (1984) reported similar re-

 Table 1

 Effects of female age and mating status on fecundity of Trichogramma minutum.

Status	Parasitized eggs per female ± SE			
	1st day	2nd & 3rd day	4th & 5th day	Avg total
Mated	64.0±3.0 a <sup>1</sup> (59) <sup>2</sup>	16.9±1.8 b (47)	3.7±0.9 b (3)	77.6±3.2 a (59)
Unmated	47.5±2.9 b (59)	35.1±2.2 a (55)	14.4±4.0 a (5)	81.4±4.0 a (59)

<sup>1.</sup> Values in the same column followed by the same letters are not significantly different at the 5% level of Duncan's multiple range test.

<sup>2.</sup> Numbers in parentheses represent replicates.

sults for *T. minutum* but Lund (1938) found that the total fecundity of unmated female *T. evanescens* was significantly higher than that of mated females.

The total fecundity for *T. minutum* in this study was lower than that reported by Yu *et al.* (1984), 200 eggs, and Smith and Hubbes (1986), 128 eggs. However, Peterson (1930) found that *T. minutum* deposited an average of 40.2 eggs in eggs of the oriental fruit moth, *Grapholita molesta* (Busck). These differences in fecundity may be attributable to host differences (Smith and Hubbes 1986). The relatively low fecundities reported here may also be due to lack of food for the females. Yu *et al.* (1984) found that *T. minutum* fed with honey produced 236.8 eggs, whereas unfed females deposited only 39.3 eggs.

The daily fecundity of T. minutum decreased significantly with age (mated: F = 96.16; df = 2, 106; P = 0.0001; unmated: F = 11.92; df = 2, 116; P = 0.0001), indicating that female Trichogramma had most of their eggs ready for deposition at or shortly after emergence. Mated and unmated females produced 82.4% and 58.3% of their total progeny on the first day. This pattern is similar to previous observations (Yu et al. 1984; Smith and Hubbes 1986). The fact that Trichogramma may lay a large proportion of their eggs on the first day of emergence should be taken into account when timing inundative field releases of Trichogramma in biological control programs.

Average longevity was different for mated and unmated T. minutum females,  $53.6 \pm 2.1$  (SE) vs  $59.6 \pm 2.1$  h, (F = 4.12; df = 1, 116; P = 0.0446). Yu et al. (1984) observed similar results with T. minutum reared on E. kuehniella, but Lund (1938) found no difference for T. evanescens. Mating is known to reduce female lifespan in many insects (Partridge 1986) and may also do so with T. minutum.

The longevity of *T. minutum* observed in this study with unfed female parasitoids was much shorter than that reported by Yu *et al.* (1984) or Smith and Hubbes (1986). Yu *et al.* (1984) found that *T. minutum* fed on honey lived for 612 h, whereas unfed females survived only 64 h. Starvation may be partly responsible for differences between measurement of female longevity

 Table 2

 Effects of female age and mating status on progeny emergence rate, clutch size and sex ratio of Trichogramma minutum.

	Emergence rate $(\%) \pm SE$					
Status		1st day	2nd & 3rd day	4th & 5th day		
Mated		95.7±1.1 a <sup>1</sup> (59) <sup>2</sup>	96.0±1.4 a (47)	95.7±8.3 a (3)		
Unmated		96.6±0.8 a (59)	95.2±2.0 a (55)	98.4±1.6 a (5)		
		Clutch	size			
Mated	X±SE	1.31±0.04 a (59)	1.25±0.04 a (47)	1.33±0.33 a (3)		
	Minimum	1	1	1		
	Maximum	5	4	4		
	Percentage of bo	th sexes in the clutch size of	of two: 76.7±5.4			
Unmated	X±SE	1.32±0.04 a (59)	1.28±0.04 a (55)	$1.25\pm0.30 a (5)$		
	Minimum	1	1	1		
	Maximum	5	4	4		
		Male proportion	on (%) ± SE			
Mated <sup>3</sup>		20.1±2.1 b (59)	32.5±3.0 a (47)	46.1±12.2 a (3)		

Values in the same column within each of emergence rates and clutch size followed by the same letters
are not significantly different at the 5% level of Duncan's multiple range test.

<sup>2.</sup> Numbers in parentheses represent replicates.

<sup>3.</sup> Values in this row followed by the same letters are not significantly different at the 5% level of Duncan's multiple range test.

here and those from previous studies. Differences in hosts may also have contributed to different measurements of longevity, because host species appear to have significant effects on parasitoid lifespans (Smith and Hubbes 1986).

### Clutch size, emergence rate and sex ratio of F1 progeny.

Mating status and age of female T. minutum did not significantly (P>0.05) affect emergence rate (Table 2). The emergence rate of parasitized eggs was above 95% for both mated and unmated parasitoids, higher than the 82.5% reported by Smith and Hubbes (1986). Although offspring clutch size varied from 1.25 to 1.32 offspring per parasitized egg, no significant (P>0.05) differences were found either between mated and unmated females or among maternal ages (Table 2).

The overall sex ratio of T. minutum was female-biased and significantly affected by maternal age (F = 8.31; df = 2, 106; P = 0.0004) (Table 2). Mated T. minutum produced a significantly lower proportion of male offspring on the first day of oviposition than on subsequent days. A similar increase in the proportion of male progeny with maternal age of T. minutum was reported by Houseweart et al. (1983) and Smith and Hubbes (1986).

Adult sex ratio was estimated in this study. In this haplodiploid parasitoid, adult sex ratio could reflect the initial sex ratio of the parasitoid eggs allocated to hosts at oviposition or differential larval mortality between sexes or both. We found that a single *P. saucia* egg can support up to five *T. minutum*, but this rarely occurs and average clutch size was only 1.3 offspring per parasitized host (Table 2). Furthermore, it was observed that if two *T. minutum* developed from a single cutworm egg, majority of them were one male and one female (see Table 2). Thus, differential larval mortality of *T. minutum* in this study is unlikely. Therefore, the observed adult sex ratio is probably determined by the initial sex ratio of the wasp eggs. In arrhenotokous Hymenoptera, males develop from unfertilized eggs and females from fertilized eggs. An increase in the proportion of males with maternal age may be due to depletion of sperm but further study is needed.

#### ACKNOWLEDGEMENTS

We are most grateful to Dr. J.H. Myers (University of British Columbia, Vancouver, B. C.) for allowing us to use her laboratory facilities for this study, and to Dr. M.B. Isman (University of British Columbia) for providing the eggs of *P. saucia*. The authors thank Ciba Geigy Canada Limited (Bio-Logicals) for providing the original *T. minutum* for this study. This research was funded in part by the National Research Council of Canada and the Science Council of British Columbia.

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# Assessment of sweepnet and suction sampling for evaluating pest insect populations in hay alfalfa<sup>1</sup>

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#### ABSTRACT

Insect populations in alfalfa grown for hay can be sampled using several methods. However, in a pest management program a relatively easy, quick, and reliable method of sampling is essential for making effective pest control decisions. A study was conducted to determine if two different sampling methods, sweepnet sampling and suction sampling, led to similar pest control decisions. Differences between sweepnet and D-Vac insect population estimates varied over sampling dates and years and were dependent on the insect species, their developmental stages, and abiotic factors. Our results indicate that, for many sampling dates, decisions on control of some pest insects would be similar for the two sampling methods.

Insecta, Medicago sativa, alfalfa weevil, pea aphid

#### INTRODUCTION

Economically viable, environmentally responsible pest insect management depends on regular, accurate assessments of insect populations. The correlation between estimates from samples and absolute population estimates varies with crop growth factors (Bechinski and Pedigo 1982, Saugstad *et al.* 1967), the insects being sampled (Sedivy and Kocourek 1988), wind, and air temperature (Saugstad *et al.* 1967). The sampling method used is also a source of error in estimating insect populations. The method chosen must be sufficiently accurate to identify population fluctuations, but also simple and quick enough that it can be done frequently to allow timely management decisions.

Comparisons of sampling methods have been inconclusive. In lentils, population estimates of *Lygus hesperus* Knight from absolute, D-Vac, and sweepnet sampling were similar, but nymphal numbers were lower with sweepnet sampling (Schotzko and O'Keefe, 1986a). In soybeans, Bechinski and Pedigo (1982) found that for the predators, *Nabis* spp., *Chrysopa* spp. and Coccinellidae, sweepnet sampling was superior, in terms of cost and variability, to plant shake, absolute and vacuum sampling. Vacuum sampling was the least efficient method. However, Shepard *et al.* (1974) found no significant differences among insect samples collected from soybeans by

D-Vac, sweepnet, and plant shake. The Insectavac was reported to sample more insects per unit area per volume of cotton sampled and, therefore, give a more accurate estimate of population density than did the sweepnet (Ellington *et al.* 1984). Smith *et al.* (1976), however, reported that both sweepnet and D-Vac sampling were adequate to identify population fluctuations and indicate absolute populations in cotton. In alfalfa, Sedivy and Kocourek (1988) found that D-Vac did not collect large, heavy insects such as caterpillars.

The objective of this study was to determine whether sweepnet and D-Vac sampling show